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Microbiotic Assessment of an Upflow Anaerobic/Aerobic Swine Treatment Process

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MICROBIOTIC ASSESSMENT OF AN UPFLOW ANAEROBIC/AEROBIC SWINE TREATMENT PROCESS

By

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Confined animal feeding operations (CAFOs) relating to swine and their resulting odors continues to be an issue of concern. The primary sources of odors from a CAFO include general ventilation of the confinement house, the anaerobic lagoon, and the land application of lagoon sludge. This paper focuses on lagoon wastewaters, but the results therein could have influence on the other two aforementioned areas.

An advanced upflow anaerobic/aerobic reactor system was developed to determine its impact on microbial activities that ultimately result in offensive odors. The microbial activity of SRB (sulfate-reducing bacteria) and hydrogen-sulfide production was monitored closely in each ‘zone’, as well as other parameters such as dissolved oxygen and BOD.

The results indicated a microbial physiology conducive to offensive odor production in the anaerobic zone of the pilot reactor and an aerobic microbial population in the upper zone of the pilot reactor. This aerobic zone was found to be effective in oxidizing the odorous gases created in the anaerobic zone. The overall microflora was consistent with an average magnitude of 10^8 CFU/mL. From the analysis performed, it was concluded that the microbiotic flora development and related substrate decomposition was the result of different metabolic pathways employed by the microflora rather than changes in the microbial population. In addition, the rise in pH throughout the experiment indicated the impact of the protein metabolic pathways (ammonification) over the carbohydrate metabolic pathways.

Overall, the upflow anaerobic/aerobic pilot reactor proved to be an effective method for ‘zoning’ of the microbiotic flora, and a positive impact on the modifying the compounds related to offensive odor production.

DEDICATION

I would like to dedicate this research to my entire family for their patience and love throughout this very long journey. To my mother, Mary McClain, and my in-laws, Polly and Carroll Christopher, I thank you for all your prayers and support.

Most importantly, I thank my wife, Resa, and my children Chris, Colby, and Alex for having confidence in me and the willingness to allow me to pursue this most important effort.

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CHAPTER I

INTRODUCTION

Nature of the Problem

With the ever-present concern about the environment in which we live, a great deal of discussion surrounding swine production and the environment currently exists. This issue has raised concerns for every aspect of the environment – land, air, and water.

In years past, small farmers, for the most part, raised their animals on open lands or in large confined pens. Most animals were born and raised on said farm, and eventually the animal was slaughtered for personal use, sold to a packing house for pork production, or kept for further breeding purposes.

Today, however, factors such as genetics, transportation, technology, and concern for disease have dramatically changed hog production methodology. "Concentrated Animal Feeding Operations" (CAFOs) as they are called, now exist in many states. These operations efficiently provide the pork industry with a much more consistent raw material than ever before. The genetic engineering now involved in this production gives producers a more homogeneous animal with which to work in terms of size, fat-to-lean, disease, and other important characteristics.

Change in one area often causes concerns in others. And the move from tradition farming techniques to one of mass production has created concerns. Air contamination,

wastewater, and land application of sludge are issues in which the owner/operator of a CAFO must deal.

Concentrated animal feeding operations

The hog factory of today has little in common with the traditional family farm of yesterday. The modern hog farm is a highly efficient, mechanized, mass-production operation. A single hog house may contain as many as a 1,000 hogs. Large hog farms having multiple houses may have as many as 10,000 hogs on a single farm. In the state of North Carolina there is one multiple house farm that has a capacity for 68,000 hogs (http://www.hogwatch.org/factory).

The primary reason for the increase in CAFOs can be traced back to North Carolina in 1989. At that time one of the largest producers of processed meats, Smithfield Foods, announced it planned to construct the world's largest slaughterhouse for pork. This facility would have the capability of processing over 24,000 hogs per day. At that time, North Carolina's hog growing capabilities were already at 2.57 million hogs (http://www.hogwatch.org/regs). Figure 1.1 shows the dramatic rise in hog production from that time.

Other pork producing states have also seen their share of increased CAFOs. Due to increases in efficiency and production, hog markets and packing capacity, and the regulatory climate that exists, states such as Iowa, Ohio, Missouri, and Mississippi have also seen CAFOs increase in number. But it is the enhanced efficiencies of the operations that have caused them to flourish.

Hog production and the environment

With this change in hog farming methodology and the increase in hog production, waste from the animals has also increased. An average size CAFO has about 3,700 hogs and produces approximately 38,500 pounds of feces and urine every day (http://www.hogwatch.org/enviroimpacts). This waste by-product must be collected and handled in an environmentally safe and acceptable manner with regard to air, groundwater, surface water, and the land.

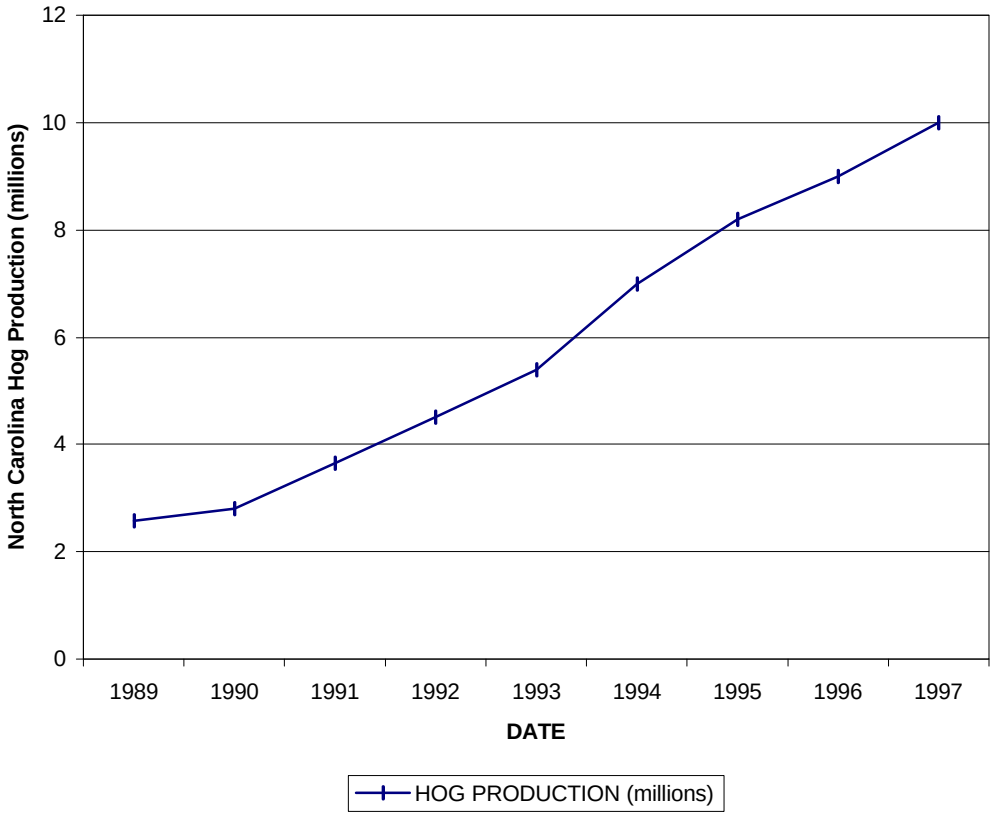

Figure 1.1 North Carolina Hog Production

In September 1998, EPA and USDA released the draft Unified National Strategy for Animal Feeding Operations. Under this draft strategy, the EPA/USDA will not require

the majority of animal feeding operations to adhere to any new federal regulations (www.epa.gov/cleanwater.afo). The 41-page document indicates that 95% of the estimated 450,000 animal feeding operations will be encouraged to voluntarily implement comprehensive nutrient management plans. The strategy also emphasizes the continuation of the voluntary approaches to implementing plans and strengthens existing voluntary programs already in place. These programs include USDA's Agriculture Research Service, Comprehensive State Research, Education, and Extension Service, and state and local programs.

These voluntary programs have resulted in the CAFO owners devising a fairly standard methodology of treatment system design. In brief, this design consists of three major components: the house, an anaerobic lagoon, and a land application system. The house, of course, houses the pigs, but allows for the feces and urine to fall through a grate system to a collection area below. This area is pre-charged with recycled lagoon water. Periodically, the entire contents of the house are emptied into the anaerobic lagoon. The anaerobic lagoon, being sized primarily on organic loading concentration, solids retention, and overflow, then treats the waste to reduce its organic loading, as measured by the biochemical oxygen demand. The material from the lagoon is then land applied through a spray system. Here, great attention is given to ensure that proper nutrient levels are maintained, and possibilities of storm runoff are minimized.

Swine and odor

While nutrient levels, groundwater contamination, and runoff are key issues of concern, the topic of odor is of much concern as well. Swine odor is a complex entity. As manure from swine decomposes, the release of many compounds may occur. Some of

these compounds include ammonia, hydrogen sulfide, organic acids, alcohols, aldehydes, amines, and mercaptans (http://www.ces.ncsu.edu/whpaper/SwineOdor.)

Aside from the compounds themselves, many other factors also play a part in the extent to which the odor is a problem. Olfactory perception varies greatly from one person to the next. Some humans can detect over ten thousand different odors, while others can only identify a small percentage of these. The psychological response to odors is also important. It is believed that odor likes or dislikes is learned behavior and therefore varies dramatically from person to person (Bundy, 1992; Donham, 1990).

The physical environment also plays a large role in the odor issue. Odors are transported by wind and ultimately diluted by atmospheric turbulence. However, many topographical features such as trees, building, hills, and hedges play major roles in how the odor is dispersed. The Gaussian plume dispersion model has been adopted worldwide as the process for quantifying the atmospheric transport of pollutants (Janni, 1994). In the approach, wind speed and temperature play a major role in the dispersion development. Other important factors such as emissions concentrations, discharge height and velocity, humidity, and specific gravity of the pollutant also dictate how and when odors may be transported (Smith, 1993). Thus, the physical environment plays a major role in determining the impact of odor on neighborhoods and communities.

To date, much work has been done to quantify odors. Some of these efforts utilize: odor sensory panels, electronic noses, gas analyzers, and gas chromatography (Kreis, 1978; Cheremisinoff, 1975). Each method has its own set of advantages and disadvantages with regard to swine odor. Perhaps the most accurate method of the four is that of the trained odor sensory panel (McGuire, 1999). The trained panel, through a

series of conditioning trials, can formulate rankings of varies types and concentrations of swine odorants, and give beneficial results to the analysis being performed.

Swine odor and the law

The federal government, through the Clean Air Act and other laws, specifically target compounds for control that may result in a hazard to human health or the environment. However, with regard to odor from concentrated animal feeding operations, there are no federal laws or regulatory requirements, thus leaving the issue of odor to the states to regulate if they so chose. Thus, there exist almost as many methods to enforce odor control as there are states. These methods range from those states that rely on the nose of the investigator to determine the extent of the odor, to those states that use odor measuring devices such as a scentometer. In some states, legislators have passed moratoria on the construction of new swine CAFOs. Additionally, local ordnances have been enacted by county governments and municipalities who implement zoning restrictions to ensure new facilities are not constructed or that proper setbacks are required.

Source of the problem

With regard to swine concentrated animal feeding operations, odors come from several sources. Sources of temporary odors include loading and unloading of animals, unloading of feedstock, and the handling and disposal of dead animals. These odors are not considered a major problem at this time. The primary odors of contention are those associated with the manure of the animals. These odors find their way into the atmosphere from three primary avenues: the general ventilation of the confinement

house, the microbial dynamics of the anaerobic lagoon, and through the spraying of lagoon sludge during the land application process.

As previously described, the common link in each of these areas is the lagoon wastewater. The current system design utilizes the lagoon wastewater as recycle water for holding the hog's excrement in suspension to facilitate removal of the material. The recycled lagoon wastewater contributes to the offensive odor in the confinement house because the anaerobic microbial flora is in a dynamic state at the time of transfer.

The swine anaerobic lagoon is a dynamic ecosystem consisting of microorganisms utilizing both organic and inorganic substrates for synthesis and respiration, but doing so in the absence of oxygen. The autotrophic and heterotrophic bacteria are continuously in a state of biochemical conversion of substrate materials to other end products. It is these end products that result in a significant odor problem. As shown in equation 1.1, organic wastes are biochemically transformed and the resulting by-products are primarily carbon dioxide, ammonia, hydrogen sulfide, and methane.

$$\text{organic wastes} \longrightarrow \text{organic acids, alcohols} \longrightarrow CO_2 + NH_3 + H_2S + CH_4 \qquad \text{Eq. 1.1}$$

The ammonia and hydrogen sulfide result in the greatest potential for offensive odors emanating from this process. Thus, as the wastewaters are sprayed into the air during land application events, and as they are recycled back into the confinement house and the vapors thereof exhausted through the general exhaust system, the opportunity for offensive odors to find their way into the atmosphere is greatly enhanced.

The anaerobic lagoon systems utilized by the majority of concentrated animal feeding operations is successful in accomplishing several objectives. The advantages and disadvantages of this type of pond system will be discussed in greater detail later in the

document, but low construction and operating costs are primary considerations. There are other types of ponds systems, however, that can be employed which offer similar treatment capabilities plus the opportunity to address odors. These pond systems include a wide variety of aerobic systems, and multiple pond systems that may use anaerobic and aerobic ponds in series. Modifications to anaerobic ponds may be considered as well. These modifications include surface aeration, spraying of an oxidizing chemical onto the lagoon surface, lagoon covers, and odor masking agents.

Objective and Scope

The functionality of anaerobic lagoon system for treating wastes of this type is well documented. Both the physical and biological performance have been extensively studied and reported. The same comment holds true for an aerobic system as well. It also has been well documented that odors can be controlled effectively where aerobic systems can be utilized.

Therefore, this study was intended to examine the impact of a modification to an anaerobic treatment system. This modification involved the introduction of oxygen into the upper layer of the anaerobic pond system. By allowing the influent materials to enter into the modified system near the bottom and discharge near the surface, coupled with the introduction of oxygen, the concept of an upflow anaerobic/aerobic system is developed. To measure the impact of such a modification, the microbial and biological elements were examined.

The scope of the study involved the simulation of an upflow anaerobic/aerobic swine treatment system using a custom built reactor that was eight feet tall. This depth, coupled

with a restricted surface area, helped simulate the stratification of environments generally observed in pond systems. Hence, the activity of the microbial system and the biological kinetics could more accurately be studied.

As previously mentioned, the gas mixtures from the anaerobic lagoon are complex and numerous. Therefore, it was important to understand the microbial activity, and seek to determine if stratification of the flora could be achieved in a single upflow system. In doing so, specific parameters, which are linked to the causation of odors, were monitored throughout the process, in both the anaerobic zone and aerobic zone of the reactor. These parameters included sulfate-reducing bacteria, hydrogen sulfide producers, carbohydrate acid producers, carbohydrate gas producers, and nitrate reducers. To determine the success of stratification of microbes, total plate counts and dissolved oxygen analyses were performed in addition to those just mentioned.

Thus, the objective of this study is to prove an effective treatment system for handling swine wastewaters can be obtained utilizing a single upflow anaerobic/aerobic system, with effective microbial and biological stratification of zones being the measurement of success.

CHAPTER II

SWINE WASTE ODOR AND ODORANTS

Odor Thresholds

As mentioned previously, odor has both an objective aspect that is measurable in concentration and duration, and a subjective aspect, such as that of offensiveness. Thus, to demonstrate the impact of the anaerobic by-products of ammonia, hydrogen sulfide, and mercaptans, it is necessary to understand their chemical properties. To quantify this impact, it is first necessary to define specific odor-related terminology. Using the established definitions found in the *Handbook of Environmental Data on Organic Chemicals*, there are at least three different odor thresholds (Verschueren, 1983):

1. the absolute odor threshold – the concentration at which 50% of an odor panel detected the odor,

2. the 50% or 100% recognition threshold – the concentration at which 50% (or 100%) of the odor panel defined the odor as being representative of the amount of the odorant being studied, and

3. the objectionability threshold – the concentration at which 50% of an odor panel finds the odor to be objectionable.

In comparing different compounds and their odor thresholds, the concept of an odor index is used. The odor index (O.I.) is a dimensionless term based upon vapor pressure and the 100% odor recognition threshold and is determined as follows:

O.I. = vapor pressure/odor recognition threshold

where, the units of vapor pressure and the 100% odor recognition threshold are in ppm, and 1 atm = 1,000,000 ppm.

The odor index is a qualitative measure of the potential of an odorant to get into the air and then to be recognized. While the odor index does not differentiate between good and bad qualities, it does provide a basis for determining which compounds would be more susceptible of posing a potential problem.

From this concept odor indexes have been categorized into three groupings:

Category I: O.I. > 1,000,000 (high odor potential)

Category II: O.I. between 100,000 and 1,000,000 (medium odor potential)

Category III: O.I. < 100,000 (low odor potential)

Table 2.1 indicates the 100% odor recognition concentrations and odor index for those compounds related to this study, and some additional common compounds. As shown from this table, the mercaptans and sulfides have a significantly higher odor index and a significantly lower 100% recognition threshold than most other compounds.

Hydrogen sulfide

Perhaps the main compound of concern with regard to odor from swine operations is hydrogen sulfide. With an odor index of 17,000,000 and a 100% recognition threshold of 1 ppm, hydrogen sulfide is a potent odorant.

Table 2.1 Threshold Odor Concentrations and Odor Index

Compound	Formula	Odor Index	100% Odor Recognition Concentration (ppm)
Isopropylmercaptan	$(CH_3)_2CHSH$	1,052,000,000	0.0002
Ethylmercaptan	CH_3CH_2SH	289,500,000	0.0020
Propylmercaptan	$CH_3CH_2CH_2SH$	263,000,000	0.0007
Methylmercaptan	CH3SH	53,300,000	0.0350
Butylmercaptan	$CH_3CH_2CH_2CH_2SH$	49,000,000	0.0008
Hydrogen Sulfide	H_2S	17,000,000	1.0000
Ethylsulfide	$(CH_3\text{-}CH_2)_2S$	14,400,000	0.0040
Methylsulfide	$(CH_3)_2S$	2,760,000	0.1000
Ammonia	NH_3	167,300	55.0
Pentane	C_5H_{12}	570	900.0
Butane	C_4H_{10}	480	5000.0
Propane	C_3H_8	425	11000.0
Heptane	C_7H_{16}	200	200.0
Octane	C_8H_{18}	100	200.0

Cox (1975) states that the most common cause of odors in wastewater systems is hydrogen sulfide, and characterizes the odor as rotten eggs, putrid, and offensive. The sulfate ion that occurs naturally in most water supply systems provides the mechanism for production of hydrogen sulfide. The sulfate ion is reduced biologically to hydrogen sulfide, H_2S. The biochemical equation for this event is shown below:

$$\text{Organic matter} + SO_4^{2-} \longrightarrow S^{2-} + H_2O + CO_2 \qquad \text{Eq. 2.1}$$

$$\text{and,}\quad S^{2-} + 2H^{+} \longrightarrow H_2S \qquad \text{Eq. 2.2}$$

Hydrogen sulfide is normally a gas, and at 20^0C and one atmosphere pressure, a liter of H_2S weighs 1.40 grams, of which 1.31 grams are sulfur. It is moderately soluble in water, as shown in Table 2.2. The proportions of H_2S and HS^- in the dissolved fraction of

sulfide fraction in water are primarily a function of pH. These proportions can be shown by the expression

$$\log [(HS^-)/(H_2S)] = pH - pK^1 \qquad \text{Eq. 2.3}$$

where pK^1 is the negative logarithm of the ionization constant. The value of pK^1 is influenced by temperature and by the ionic strength of the solution. Generally speaking, the pK^1 for hydrogen sulfide is close to 7.0. Table 2.3 shows the proportions of dissolved sulfide existing for H_2S as function of $pH-pK^1$.

Table 2.2 Solubility of Hydrogen Sulfide at a Pressure of One Standard Atmosphere

Temperature, ^0C	Solubility, mg/L as $S^=$
0	6648
5	5646
10	4810
15	4150
20	3618
25	3175

Hydrogen sulfide, fortunately, is a weak diprotic acid that spontaneously oxidizes under aerobic conditions. In the bottom layer of anaerobic pond systems, hydrogen sulfide is normally produced. As these vapors rise and come into contact with higher levels of dissolved oxygen, the hydrogen sulfide reacts with the oxygen to yield water and elemental sulfur. Equation 2.4 shows the result of the hydrogen sulfide and dissolved oxygen reaction.

$$H_2S + 1/2O_2 \longrightarrow H_2O + S^0 \qquad \text{Eq. 2.4}$$

However, most anaerobic pond systems do not maintain high enough dissolved oxygen levels near their surface to produce this reaction. This fact provides the support, in part, for the upflow anaerobic/aerobic treatment concept.

Table 2.3 Proportions of Dissolved Sulfide Present as Hydrogen Sulfide

pH – pK	pH if pK = 7.0	Proportions of hydrogen-sulfide	pH – pK	pH if pK = 7.0	Proportion of hydrogen-sulfide
-2.0	5.0	0.990	0.4	7.4	0.280
-1.8	5.2	0.980	0.5	7.5	0.240
-1.6	5.4	0.975	0.6	7.6	0.200
-1.4	5.6	0.960	0.7	7.7	0.170
-1.2	5.8	0.940	0.8	7.8	0.140
-1.0	6.0	0.910	0.9	7.9	0.110
-0.9	6.1	0.890	1.0	8.0	0.091
-0.8	6.2	0.860	1.1	8.1	0.074
-0.7	6.3	0.830	1.2	8.2	0.059
-0.6	6.4	0.800	1.3	8.3	0.048
-0.5	6.5	0.760	1.4	8.4	0.039
-0.4	6.6	0.720	1.5	8.5	0.031
-0.3	6.7	0.670	1.6	8.6	0.025
-0.2	6.8	0.610	1.7	8.7	0.020
-0.1	6.9	0.560	1.8	8.8	0.016
0.0	7.0	0.500	1.9	8.9	0.013
0.1	7.1	0.440	2.0	9.0	0.010
0.2	7.2	0.390	2.5	9.5	0.003
0.3	7.3	0.330	3.0	10.0	0.001

Ammonia

In swine wastewaters, ammonia originates from the hydrolysis of urea in the urine. With an odor index of 167,000 and a 100% recognition threshold of 55 ppm, ammonia is a significant odorant. Cox (1975) describes the odor from ammonia vapors as very

pungent, resembling dry urine. Ammonia odors can be formed aerobically as well as anaerobically, so the control strategies recommended for anaerobic odors may not apply (Richard, 1996). The microorganisms are very efficient at utilizing nitrogen when that is the limiting nutrient. The smell of ammonia is an indicator that nitrogen is in excess, and carbon/energy is limiting instead.

Another factor affecting the magnitude of ammonia volatilization is pH (Shakhashiri, 2000). NH_3 (gaseous ammonia) and NH_4^+ (aqueous ammonium ion) are in equilibrium at a pH of about 9. Higher pH values will, therefore, force more NH_4^+ into a gaseous state. Thus, ammonia is rarely noticed if the pH is acidic. This pH equilibrium curve is shown in figure 2-1 and the quantitative relationship is shown by equation 2.4.

$$K_b = (NH_4^+)(OH^-)/(NH_3) = 1.8 \times 10^{-5} \text{ at } 25^0C \qquad \text{Eq. 2.4}$$

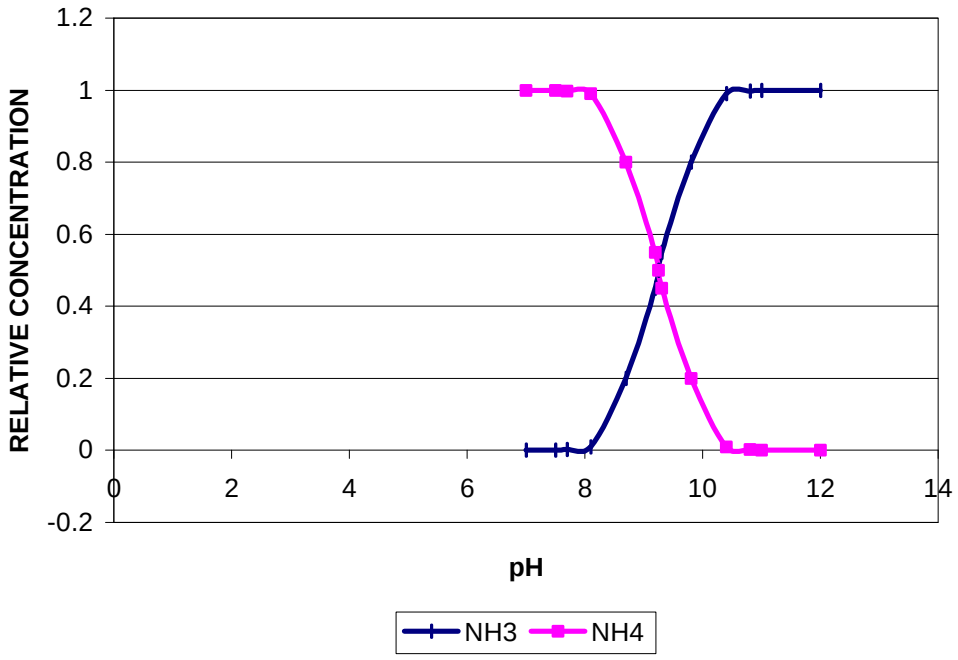

Figure 2.1. Relative Concentrations of NH_3 and NH_4

In order for ammonia to be converted to nitrite and then to nitrate, the autotrophic bacteria of the genera *Nitrosomonas* and *Nitrobacter* are required. However, in many anaerobic lagoon systems where no cover is employed, there exist aerobes at or near the surface, facultative anaerobes slightly below the surface extending to the sludge blanket near the bottom, and an anaerobic zone in the bottom.

Thus, in systems where the dissolved oxygen concentration is above 1 mg/l, nitrification can occur. Bacteria of the genera *Nitrosomonas* and *Nitrobacter* usually exist in sufficient quantities to drive ammonia oxidation provided cell residence time is high enough. This process is a two-step event that is described as follows:

$$NH_4^+ + 3/2O_2 \longrightarrow NO_2^- + 2H^+ + H_2O \quad (\textit{Nitrosomonas}) \quad \text{Eq. 2.5}$$

$$NO_2^- + 1/2O_2 \longrightarrow NO_3^- \quad (\textit{Nitrobacter}) \quad \text{Eq. 2.6}$$

Thus, the overall energy reaction is:

$$NH_4^+ + 2O_2 \longrightarrow NO_3^- + 2H^+ + H_2O \quad \text{Eq. 2.7}$$

In addition, some of the ammonia is assimilated into cell tissue. This reaction is given by:

$$4CO_2 + HCO_3^- + NH_4^+ + H_2O \longrightarrow C_5H_7O_2N + 5O_2 \quad \text{Eq. 2.8}$$

Thus, the total overall reaction including both energy and cell synthesis is given by:

$$NH_4^+ + 1.83O_2 + 1.98HCO_3^- \longrightarrow 021C_5H_7O_2N + .98NO_3^- + 1.041H_2O + 1.88H_2CO_3 \quad \text{Eq. 2.9}$$

Therefore, from this equation, it takes approximately 2.3 mg O_2/mg ammonia nitrogen to oxidize ammonia to nitrate.

To move the process further to completion requires a third step, which must take place under anoxic conditions. In the denitrifying process, the dissolved oxygen concentration is extremely important. The presence of dissolved oxygen will suppress the enzyme system needed for denitrification. Several heterotrophs are capable of reducing nitrate to nitrogen gas. Some of these include organisms of the genera *Enterobacter, Bacillus,* and *Pseudomonas*. The reactions for nitrate reduction are given by:

$$NO_3^- \longrightarrow NO_2^- \longrightarrow NO \longrightarrow N_2O \longrightarrow N_2 \qquad \text{Eq. 2.10}$$

Mercaptans

The most important thing to know about mercaptans is that they stink (www.cng.com). With odor indexes ranging from 2,760,000 for methylsulfide to 1,052,000,000 for isopropylmercaptan, there is no doubt that mercaptans, if present at all, are an odor problem. All mercaptans contain sulfur. As noted in Table 2.1, the 100% Odor Recognition Concentration for the aforementioned mercaptans are 0.1 parts per million and 0.2 parts per billion, respectively. Thus, a small amount of mercaptans when combined with other suspected odorous compounds as found in swine wastewaters, can be offensive.

CHAPTER III

WASTE TREATMENT POND SYSTEMS

The confined animal feeding operations for growing hogs use, for the most part, anaerobic lagoon systems for treating their wastewaters. While many other options exist, the rationale for choosing this method of treatment has to do with its effectiveness and low operational costs. To further explain how the CAFO designers came to decide upon this type of treatment system, it is necessary to take a brief look at the history of wastewater treatment.

Dating back to the early 1800's disposal of human waste has been a recognized problem. However, at that time it was up to the individual to handle his or her waste. In Europe, where dense populations of people existed, much the waste made its way to cesspools intended specifically for holding the waste materials. Overflows, however, permitted the wastes to flow into the public streets and rivers. Many rivers and streams were subsequently transformed from pristine sources of water to stench-ridden pools that were sources of disease.

It was not until the early 1900's that significant progress was made in the area wastewater treatment. The realization that human excrement was closely associated with the transmission of feared and lethal diseases finally brought about significant change (Oswald, 1994). Epidemics of cholera and typhoid fever instigated the application of the

principals of microbiology. And as more and more developing countries embraced the scientific communities' recommendations, concerted efforts were made to eliminate unsightly and unsanitary conditions by the collection of sewage wastes. In the beginning, these materials were treated only by the prevention of floatable and settleable solids from entering the streams and rivers. Thus, these methods did not remove significant numbers of microbes or soluble organic substances.

The first real treatment system to evolve was the septic tank. In the 1860's it was noticed that waste materials that stayed in seepage pits for a brief period of time, and then overflowed, were less odorous than otherwise. It was further noticed that these materials in the seepage pits went through a gas-producing fermentation process. In 1878 Alexander Muller applied for patent rights for a process in which wastewaters were biologically treated in tanks in which air was excluded (Oswald, 1994). Thus, the first simple septic tank was born.

But as communities grew, the need for larger systems grew as well. This fact brought about the concept of settling in ponds to remove the solids, followed by land irrigation. This concept proved satisfactory in some areas where the soil conditions were favorable for absorbing the nutrients, but not so favorable in areas where dense soils were found. In addition, excessive rainfall caused problems with runoff into streams and rivers. But the biggest problem was with the manpower needed to apply the solids to the fields. It is believed that few of the sewage farms were actually profitable, and therefore, still other methods of disposal were sought.

As time went on, research was done on a wide variety of methods including chemical treatment, physical treatment, biological systems using underdrainage, and biofiltration.

As the microbiology of each of these was better understood, progress was being made toward a better understanding of the advantages and disadvantages of each.

Caldwell (1946) first outlined specific design criteria for ponds. In his work, Caldwell emphasized oxidation and suggested that ponds not exceed four to five feet in depth so that wind would mix the dissolved oxygen that had been produced by algae. Some years later, however, it became clear that these systems were discharging effluents which still had algae remaining in them, and therefore, were exerting an oxygen demand equal to or greater than the original wastewater influents. This fact led to additional research and in the 1960's, Oswald et al. (1963) suggested that ponding systems that had anaerobic zones in which methane fermentation could occur gave better results than the shallower ponds.

Meanwhile, other work continued on aerobic systems for both attached and suspended growth systems. Activated sludge systems, oxidation ditches, and trickling filters became familiar aerobic designs for many wastewater engineers. Today many wastewater treatment systems incorporate anaerobic and aerobic treatment in series in order to satisfy water quality objectives.

The following discussion of the three major ponds systems individually helps to understand the biological and biochemical results of each, and leads us to understand how they might be combined into an upflow system.

Anaerobic pond systems

In the anaerobic pond system, there is no free oxygen or mixing and it is therefore anaerobic throughout its depth. In the anaerobic digestion process, organic waste materials are converted, in the absence of oxygen, to other end products. These end

products consist primarily of methane and carbon dioxide, but usually also contain small quantities of hydrogen sulfide, hydrogen, organic acids, and cell tissues (Glysson et al., 1985). The process comprises three stages (Reynolds, 1982):

1. organic materials are transformed to organic acids,
2. organic acids are reduced, in part, to methane and CO_2, and
3. carbon dioxide is reduced with water to form methane.

The first step in the process involves the enzyme-assisted transformation or hydrolysis of higher-molecular-mass compounds into compounds suitable for use as source of energy and cell carbon. The second step, called acidogenesis, converts the now smaller molecular units into short-chain organic acids such as acetic acid (CH_3COOH), propionic acid (CH_3CH_2COOH), and butyric acid ($CH_3CH_2CH_2COOH$). A heterogeneous population of facultative and anaerobic bacteria is responsible for these hydrolytic and oxidation reactions. In the acid fermentation stage no COD reduction occurs since the primary activity is the conversion of complex organic molecules to short-chain organic acids.

In the methane fermentation stage, methanogenic microoganisms, which are strictly anaerobic, convert the longer chain acids to methane, carbon dioxide, and organic acids having a shorter carbon chain (Ramalho, 1983). The acid molecules are broken down yielding acetic acid, which is then converted to carbon dioxide and methane as shown in equation 3.1.

$$CH_3COOH \xrightarrow{\text{methanogenic bacteria}} CO_2 + CH_4 \qquad \text{Eq. 3.1}$$

The group of facultative and anaerobic bacteria that is responsible for the acid fermentation stage has a much faster rate of growth than the methanogenic bacteria. As a

result, the acid fermentation stage is relatively rapid and the methane fermentation stage is the rate-controlling step in the anaerobic process. Thus, detention time for methane microorganisms must be adequate or they will cease to exist. For an anaerobic lagoon system, this detention time is 2 to 20 days, with the optimum pH of 6.8 to 7.4 (Oswald, 1994).

The loading of an anaerobic lagoon is critical in that anaerobic conditions must be maintained at all times. If influent wastewater flows are allowed to enter above the lagoon surface, oxygen may be entrained in the liquid and result in area of unpredictable treatment and odors. It is also desirable to have consistent influent flow rates and organic loadings. Organic loadings normally range between 250 and 4000 pounds of BOD_5 per acre per day, with BOD_5 removal efficiencies between 50% and 80%. Anaerobic lagoon depths usually range between 8 feet and 15 feet, but greater depths are not uncommon.

Aerobic pond systems

The aerated lagoon system evolved from facultative stabilization ponds when aerators were installed in an effort to reduce odors from lagoon systems which were overloaded with organic materials.

In the aerobic lagoon system, the objective is similar to that of the anaerobic lagoon, that is, to stabilize the waste (Mitchell, 1974). In this system, as organic wastes are introduced, oxygen is added through mechanical means and aerobic digestion takes place, with the end products being carbon dioxide and water. The mechanical aeration not only serves to provide an adequate supply of oxygen, but also to provide complete mixing. The digestion process is a two-step process as shown in equations 3.2 and 3.3 below:

$$COHNS + O_2 + \text{nutrients} \xrightarrow{\text{bacteria}} CO_2 + NH_3 + C_5H_7O_2N + \text{other end products}$$

Eq. 3.2

$$C_5H_7O_2N + 5O_2 \xrightarrow{\text{bacteria}} 5CO_2 + 2H_20 + NH_3 + \text{energy}$$ Eq. 3.3

In these equations, COHNS represents the organic waste matter in the wastewater and $C_5H_7O_2N$ represents new bacterial cells. As shown, equation 3.3 represents the endogenous respiration for the bacteria, and results in simple and stable end products.

The bacteria are the most important microorganisms in the system because they are responsible for the decomposition of the organic matter in the waste. In performing their job, part of the substrate is used for cellular material and part for energy. The substrate used to produce new microoganisms, called synthesis, also results in an increase in biomass. The substrate used for energy is for cell maintenance and mobility.

Because the substrate is continuously utilized for synthesis and cell maintenance, the concentration of the organic material is ultimately depleted. If the source of organic material is allowed to become exhausted, the bacteria will enter into the endogenous respiration phase. This phase is given by the equation 3.4 below:

$$C_5H_7O_2N + 5O_2 \longrightarrow 5CO_2 + NH_3 + 2H_2O$$ Eq. 3.4

Aerated lagoons typically have depths from 4 to 12 feet and oxygen is added to the wastewater by surface, turbine, or diffused methods. Detention times are usually less than three days.

Aerobic systems without mixing are also used to stabilize waste organic materials. Here, shallow basins are constructed that utilize algae and aerobic bacteria to perform their function. Oxygen enters the system by means of atmospheric diffusion, and through that produced by the algae. In this type of system, a cyclic-symbiotic relationship exists.

The oxygen released by the algae through the photosynthesis process is used by the bacteria in the aerobic degradation of organic material (Fry et al., 1992). The nutrients and carbon dioxide released in this degradation are then used by the algae. Temperature and solar radiation also play an important factors in the success of this type of pond system.

The BOD conversion is quite high at up to 95% of substrate. The downside is, however, that the effluent may contain a high BOD loading due to the carryover of algae and bacteria in the effluent.

Stabilization pond systems

A stabilization pond is a shallow body of wastewater that has no aeration equipment. Also called an oxidation pond, it is very popular in small communities and industries such as oil refineries and food related process industries (Ramalho, 1983; Pelczar, Chan, and Kreig, 1986; Peppler and Perlman, 1979). Stabilization ponds in which the upper layers are aerobic and the lower layers are anaerobic are referred to as facultative ponds.

Oxygen needs for these ponds are provided by natural surface aeration and by algae that produce oxygen by photosynthesis. The oxygen released by the algae as a result of the photosynthesis is utilized by bacteria for aerobic degradation of organic matter. The products of the organic matter degradation are carbon dioxide, ammonia, and phosphates. The algae use these products to produce new algae. Equation 3.5 shows the ideal photosynthetic equation.

$$CO_2 + 2H_2O \longrightarrow (CH_2O)_X + O_2 + H_2O \qquad \text{Eq. 3.5}$$

Here, CH_2O is regarded as the organic matter fixed in plant material. Oxygen is produced only as a result of a net gain in $(CH_2O)_X$. And, since the O_2 generated has been

proven to come entirely from the water on the left-hand side of the equation, there is essentially an infinite supply of oxygen with which to oxidize incoming waste (Oswald, 1994). An analysis of the material can then be approximated as shown in equation 3.6.

$$106CO_2 + 236H_2O + 16NH_4^+ + HPO_4 \xrightarrow{\text{light + algae}} C_{106}H_{181}O_{45}N_{16}P + 118O_2 + 171H_2O + 14H^+ \qquad \text{Eq. 3.6}$$

Thus, stoichiometrically the ratio of oxygen released to algal cell material grown is 1.55.

The process of producing the oxygen by photosynthesis is cyclic. During the day, in the presence of sunlight, photosynthesis takes place and oxygen is produced. During the daylight hours, some of the oxygen produced is utilized for respiration purposes; however, a substantial surplus of oxygen may prevail during the day. At night, when there is no oxygen production, algae and bacteria use oxygen, thereby lead to a depletion in dissolved oxygen. Also, during the night, the pH drops because the released carbon dioxide decreases. During the day the ammonia resulting from the degradation of nitrogenous organic compounds contribute to and increase the pH. Thus, a stabilization pond may be basic during the day, and acidic at night. In addition, where wastewaters have low initial alkalinities, high pH conditions can occur because the algae utilize the available carbon dioxide in photosynthesis activity (Tchobanoglous and Burton, 1991).

The discussion of pH is extremely important with regard to the issue of odor as well. Oswald (1994) suggests that the organic acids which are formed by the facultative heterotrophs in the anaerobic layer of the pond may be accompanied by a decrease in pH in systems that are not well buffered. A decrease in pH below 7.5 will be often be accompanied by a release of hydrogen sulfide (H_2S) to the air. Altogether a high pH

prevents the release of H_2S. If the HS^- is in the ionic form it remains in the water. Equation 3.7 shows this relationship.

$$H_2S \underset{\text{basic}}{\overset{\text{acidic}}{\rightleftharpoons}} HS^- + H^+ \qquad \text{Eq. 3.7}$$

The depth of oxygen penetration is also an important characteristic of the stabilization pond. According to Oswald (1994), the depth of oxygen penetrations is related to the loading. The greater the loading, the shallower the depth of oxygen penetration since the oxygen demand is higher.

Stabilization ponds may be used in parallel or in series to achieve specific objectives (Tchobanoglous and Burton, 1991). Where high levels of organic loading are found and biochemical oxygen demand levels are high, series operations are beneficial.

CAFO ponds

To satisfy the requirements of the CAFO wastewater needs, several different methods to could be employed. The method chosen, however, is most commonly an anaerobic lagoon system. This type of pond system has several advantages over its competition. These advantages are found in the areas of:

- Minimum design and construction time,
- Consistency in treatment performance,
- Lack of chemicals required,
- Low power consumption,
- Limited manpower required to operate,
- Low first cost to construct,
- No sludge disposal in daily operations, and

- Simplicity of operation.

However, the anaerobic lagoon system has disadvantages as well. These disadvantages include:

- Sludge disposal concerns,
- Fluctuations in lagoon levels due to evaporation and leakage,
- Overflow from stormwater events, and
- Offensive off-gas production.

If properly designed, monitored, and maintained the first three disadvantages can be properly handled. The issue of offensive off-gas production, however, requires additional attention. Thus, as previously stated, it is the intent of this research to attempt to address this disadvantage by analyzing a single upflow anaerobic and aerobic treatment system.

CHAPTER IV

MICROBIAL ELECTRON TRANSPORT AND MOTILITY

Chemiosmotic theory

In the analysis of any biological treatment system, the ability for specific microorganisms to seek environments in which they may adapt and flourish is critical if the system is to perform as desired. To have a diverse ecosystem in this treatment process, specific nutrients and different levels of oxygen are required. It is essential, therefore, for the microorganisms to be motile in their efforts to seek more favorable environments. The following discussion explains the basics of this motile activity.

According to the theoretical ideas of Mitchell, the concept of membrane bioenergetics is well understood (White, 2000). His work provides the basis for understanding how bacteria function. Similar to a battery that maintains a potential difference between its positive and negative poles for current flow of electrons, the cell membranes of the bacteria produce a proton potential difference between outside and inside. The chemiosmotic theory, as it is called, states that energy-transducing membranes pump protons across the membrane, thereby generating an electrochemical gradient of protons across the membrane that can be used for doing work when the protons return across the membrane to the lower potential.

These proton conductors are transmembrane proteins. Some membrane protons are solute transporters, others synthesize ATP, and others drive flagella rotation. The amount of electrochemical work able to be performed when an ion crosses a membrane is a function the membrane potential and the difference in concentration between the solutions separated by the membrane. The term 'proton motive force' associated with this concept is defined as potential energy in the electrochemical proton gradient. Thus, when cells move toward the lower electrochemical proton gradient the proton motive force gives up energy and work is done.

Electron carriers and electrode potential

By coupling the flow of electrons through membranes to the creation of an electrochemical proton gradient, energy is generated for growth-related processes. This flow of electrons via electron carriers is known as respiration. If the terminal electron acceptor is oxygen, the electron flow is called aerobic respiration. If it is not oxygen, then it is called anaerobic respiration. There is a continuous flow of electrons through electron carriers in bacterial cell membranes from low potential electron donors to high potential electron acceptors. The electron acceptors can be oxygen or some other inorganic acceptor such as nitrate or sulfate. Thus, there is oxygen respiration, nitrate respiration, and sulfate respiration.

Each of the electron carriers has a different electrode potential, and the electrons are transferred sequentially to a carrier of a higher potential. Table 4.1 below shows some of the standard potentials of electron donor and acceptors for a pH of 7 (White, 2000). The tendency of a molecule to accept an electron from another molecule is given by its electrode potential, E. The more positive the electrode potential, the more oxidation is

occurring. The more negative the electrode potential, the more reduction is occurring. Schulz and Barnes (1990) contend that oxidation/reduction potential is an indicator of the presence of odorous compounds in wastewaters. Their research indicated that the redox potential was superior to dissolved oxygen as a parameter in that it was better able to identify the existence of reducing conditions which are known to give rise to the generation of odorous compounds, including volatile fatty acids. Previous works by Barnes (Barnes et al., 1985) indicated that swine wastewaters are not odorous if maintained at a redox potential of at least 40 mV with respect to the standard hydrogen electrode (E_h).

Table 4.1 Standard Potentials of Electron Donors and Acceptors

COUPLE	E(mV)
O_2/H_2O	+815
NO_3^-/NO_2^-	+421
Pyruvate/lactate	-185
S^0/H_2S	-270
H^+/H_2	-410
CO_2/formate	-432

Flagella and motility

As mentioned previously, some of the proton potential is used to drive flagella rotation. To assist the bacteria in movement, swimming bacteria have one or several flagella. These flagella are organelles that protrude from the cell surface and rotate like a propeller. The rotation of the flagella motor functions as an electrochemical machine. The energy to drive the motor comes from a current of protons that moves down a proton potential gradient through the flagella motor from the outside of the membrane to the inside. The passage of the proton turns the motor in a way that causes the filament to rotate and, in turn, propels the bacteria through the media.

The bacteria are, therefore, able to swim toward more favorable environments. Each type of bacteria then may seek its own preferred location with regard to nutrients, light, and electron acceptors. In addition, the bacteria are also capable of moving to avoid undesirable environments, such as toxicity.

Terminal electron accepting reactions

The last step in the flow of electrons through the microbial food chain is called the terminal electron accepting reaction. The last compound that is reduced is the terminal electron acceptor. This process is an indicator of the nature of the microbial community in the overall flora.

Table 4.2 shows the most commonly available terminal electron acceptors (Verschueren, 1983). Oxygen is the electron acceptor that provides the greatest energy yield. Thus, when it is present, aerobic metabolism will dominate. After oxygen, nitrate is the next electron acceptor in the progression sequence. Iron, manganese, and sulfate

sequentially follow nitrate as electron acceptors. When the other electron acceptors have been depleted, carbon dioxide becomes the terminal electron acceptor.

When oxygen is the terminal electron acceptor, there will be no opportunity for iron reduction, sulfate reduction, or methanogenesis because the oxygen is toxic to the obligate anaerobic processes. Only at very low oxygen concentrations is the potential for nitrate competition with oxygen for available electrons.

Table 4.2 Terminal Electron Acceptors

Electron acceptor	Reduced product
O_2	H_2O
NO^{3-}	N_2
Fe(III)	Fe(II)
Mn(IV)	Mn(II)
SO_4^{2-}	S^{2-}
CO_2	CH_4

When nitrate is an electron acceptor, it is reduced either to nitrogen gas by denitrification or to ammonium ions by dissimilatory nitrate reduction. Nitrate reduction to ammonium ions appears to be the preferred pathway when the electron supply greatly exceeds the amount of available nitrate (White, 2000).

CHAPTER V

ANAEROBIC / AEROBIC SYSTEM FLORA

Pond systems

A great deal of pond system research and development has been done by Dr. William Oswald. In his syllabus on Advanced Integrated Pond Systems, Oswald (1994) discusses pond systems and classifies them by:

1) oxygen resources,
2) major microbiological activity,
3) sequence,
4) overflow,
5) and, integrated ponds.

Specifically, with respect to the classification regarding major microbiological activity, Oswald describes the systems as follows:

a) Oxidation pond – a pond in which biological oxidation with molecular oxygen is the primary mode of waste stabilization. The primary source of oxygen is from photosynthesis, but may also come from mechanical aeration. The primary end products are carbon dioxide, waster, and ammonia.

b) Acid fermentation pond – a pond in which heterotrophic fermentation predominates. Brought about by excessive loading, these acidic ponds are extremely odorous, and therefore, undesirable. The end products are organic acids, carbon dioxide, hydrogen sulfide, and volatile acids.

c) Methane fermentation pond – as the name implies a pond in which methane fermentation predominates. Oswald states that ponds operating under these conditions can accept heavy BOD loadings without objectionable odors due to their neutral or alkaline pH and buffering capacity. This fact prevents low pH conditions and hydrogen sulfide emissions. The end products of the methane fermentation pond are carbon dioxide, methane gas, and nitrogen gas.

d) Algae pond – a pond in which algal biomass predominates instead of bacteria. Also termed High Rate Pond, they can accept high organic loads, and may have a BOD removal efficiency of over 90% due to the oxygen production of the algae. The major end products are algal cells and dissolved oxygen.

Table 5.1 provides a summary of characteristics and environmental requirements of the major biological reactions for each type of pond.

Much of Oswald's work involved the research of pond systems in an effort to make them much more reliable and economical. This work resulted in the concept of an Advanced Integrated Wastewater Ponding System, or AIWPS. This concept involves the construction of four ponds in series. A cross section of these ponds is shown in Figure 5.1.

Table 5.1 Summary of Characteristics and Environmental Requirements of the Major Biological Reactions in Waste Disposal Ponds (Oswald, 1994)

Biological Reactions	Characteristics				
	Organisms	Usual Substrates	Major Products	Time Required (days)	Odors Produced
Aerobic Oxidation	Aerobic Bacteria	Carbohydrates, Proteins	$CO_2 + NH_3$	2 – 3	None
Photo-Synthetic Oxygenation	Micro-Algae	CO_2, NH_3	Oxygen, Algae	3 – 4	None
Acid Formation	Facultative Heterotrophs	Carbohydrates, Proteins, Fats	Organic Acids	5 – 10	H_2S, Organic Acid
Methane Fermentation	Methane Producers	Organic Acids	CH_4, CO_2, H_2	20 – 40	H_2S

Biological Reactions	Environmental Factors					
	Temp ^0C	Temp Range ^0C, permissible input	Mechanical Oxygen	pH	Light	Toxic Compounds
Aerobic Oxidation	0 – 40	15 – 30	Required	7.0 – 9.0	Not Req'd	Cr^{+++}, NH_4^+
Photo-Synthetic Oxygenation	4 – 40	15 – 25	Required under certain conditions	6.5 – 10.5	Req'd	Ca^{++},Cl_2 Cr^{+++}
Acid Formation	0 – 50	10 – 40	Required under certain conditions	4.5 – 8.5	Not Req'd	Cr^{+++}, Cl_2
Methane Fermentation	6 – 30	14 – 30	Must be excluded	6.5 – 8.0	Not Req'd	O_2,NH_4^+, Na,Ca

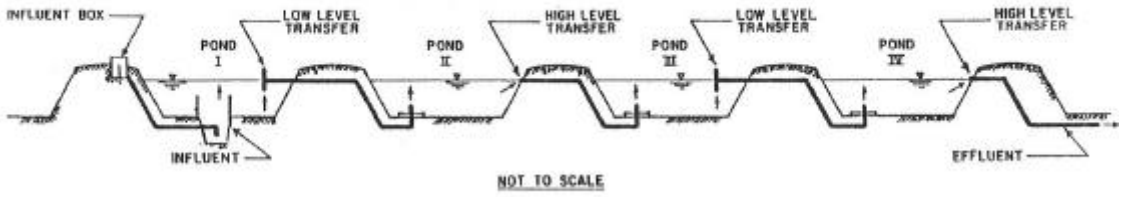

Figure 5.1 Typical Arrangement of Transfer Structures to Avoid Short Circuit of Influent to Effluent Due to Thermal Density Differences Between Influent and Pond Contents. Arrangement for Four Ponds. (Oswald, 1994)

The first of the ponds is an Advanced Facultative Pond (AFP). This pond is aerobic on the surface and anaerobic near the bottom. In the AFP, as opposed to conventional stabilization ponds, sedimentation and methane fermentation pits are constructed to avoid the intrusion of dissolved oxygen. The raw wastewater is introduced near the bottom of these pits, and most of the settleable solids remain within the pits. Alternative configurations for AFP inlets into the fermentation pits are shown in Figure 5.2. A well-designed AFP will remove 60% of the influent BOD and almost all suspended solids. Table 5.2 shows the organic loading in the AFP as function of temperature and solids.

In addition, for lightly loaded systems, the AFP will yield a dissolved oxygen profile similar to that of the stabilization pond. This profile is shown graphically versus depth in Figure 5.3. As shown, at a depth of three feet and greater, the dissolved oxygen drops dramatically until it approaches zero at approximately 5.5 feet.

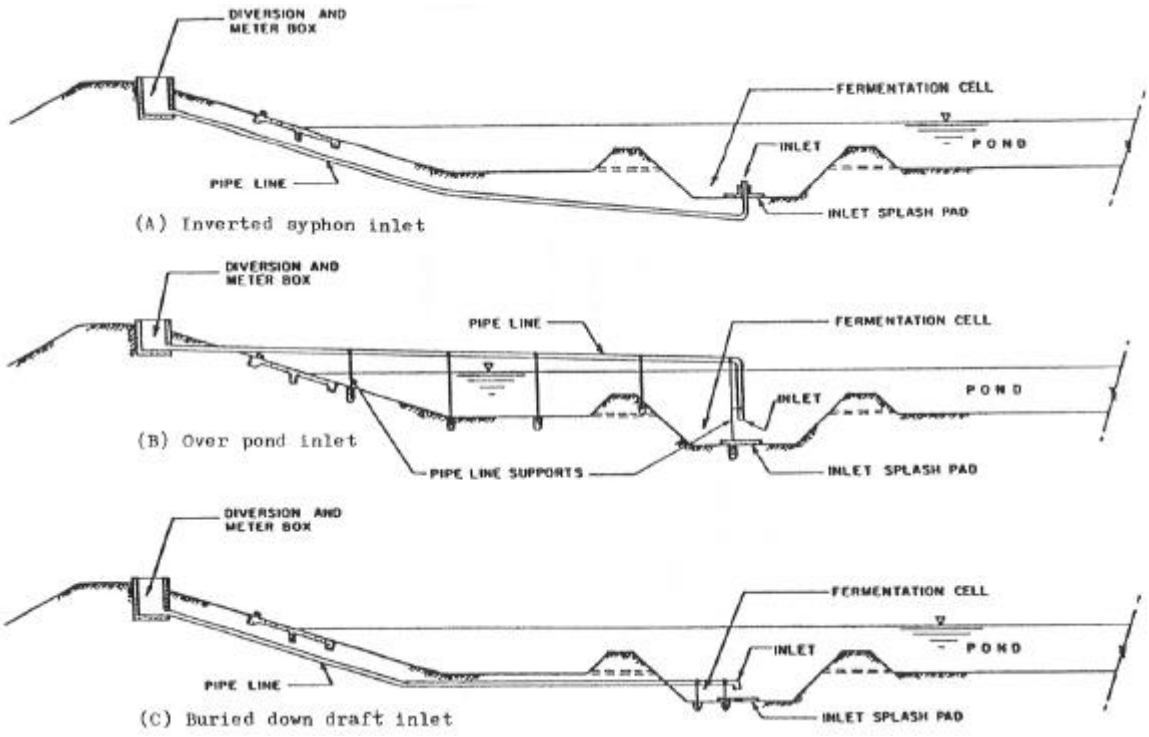

Figure 5.2 Alternative Primary Pond Inlets Into Fermentation Pits (Oswald, 1994)

The second pond of the AIWPS is a Secondary Facultative Pond (SFP) or also called a High Rate Pond (HRP). A HRP will include an aeration system that will mix and generate 100 to 300 pounds of dissolved oxygen per acre. The system also generates 25 to 200 pounds of algae biomass. If paddle wheel aerators are used as means of mixing, approximately one-tenth of a kilowatt-hour in paddle wheel mixing energy is required to produce on kilogram of algae. And, this kilogram of algae during growth will release about 1.5 kilograms of oxygen. Thus, the oxygenation efficiency of the HRP is between 10 to 15 kilograms of dissolved oxygen per kilowatt-hour. And, since the oxygen transfer rate of mechanical aeration is approximately one kilowatt hour per kilogram of oxygen transferred, algal HRP's can be much more economical than mechanical aeration.

Table 5.2 Maximum Permissible Organic Loading in Anaerobic Pits of Facultative Ponds or in Anaerobic Ponds as a Function of Temperature and Solids (Oswald, 1994)

% Solids	Temperature, ^0F / ^0C													
	40^0 / 4.5^0		50^0 / 10^0		60^0 / 15.8^0		70^0 / 21^0		80^0 / 27^0		90^0 / 32.5^0		100^0 / 38^0	
	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day	lb/ft^3/day	kg/m^3/day
1	0.0040	0.0064	0.0080	0.1280	0.0100	0.1600	0.0130	0.0280	0.0160	0.2560	0.0210	0.3370	0.0250	0.4010
2	0.0080	0.1280	0.0160	0.2560	0.0200	0.3210	0.0260	0.4170	0.0320	0.5140	0.0420	0.6740	0.0500	0.8020
3	0.0120	0.1930	0.0240	0.3850	0.0300	0.4180	0.0390	0.6260	0.0480	0.7700	0.0630	1.0100	0.0750	1.2000
4	0.0160	0.2570	0.0320	0.5140	0.0400	0.6420	0.0520	0.8340	0.0640	1.0300	0.0840	1.3500	0.1000	1.6000
5	0.0200	0.3210	0.0400	0.6400	0.0500	0.8030	0.0650	1.0400	0.0800	1.2800	0.1050	1.6800	0.1250	2.0000
6	0.0240	0.3850	0.0480	0.7700	0.0600	0.9630	0.0780	1.2500	0.0960	1.5400	0.1260	2.0220	0.1500	2.4100
7	0.0280	0.4490	0.0560	0.8980	0.0700	1.1200	0.0910	1.4600	0.1100	1.7600	0.1470	2.3600	0.1750	2.8100
8	0.0320	0.5140	0.0640	1.0200	0.0800	1.2800	0.1040	1.6700	0.1280	2.0500	0.1680	2.6400	0.2000	3.2100

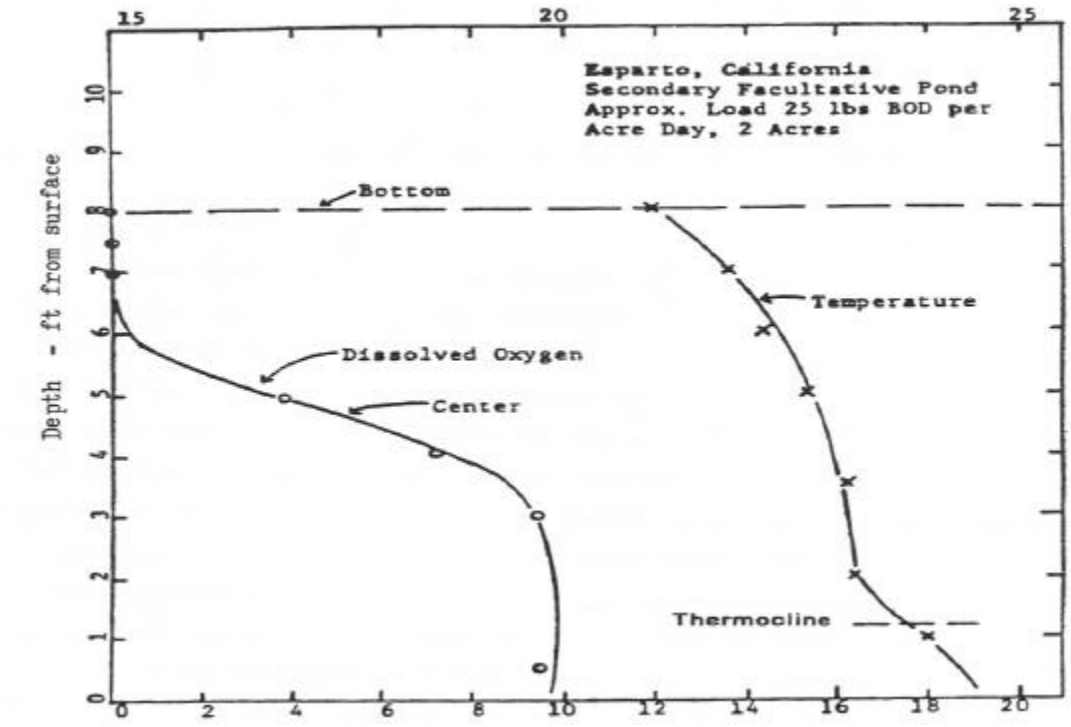

Figure 5.3. Dissolved Oxygen, mg/L (Oswald, 1994)

The third pond in the AIWPS is simply an Algal Settling Pond (ASP). However, Oswald points out that as uses for waste-grown algae may increase, in which case, the algae could be harvested and marketed.

The final pond in the AIWPS is a disinfecting pond. However, rather than chlorination, storage for 10 to 20 days in a deep maturation pond will provide adequate kill of pathogenic microorganisms of human origin.

Figure 5.4 shows the comparative relative activity rate versus pH for each of the major microbial processes described. Of interest are the width of variation for organic

acid formation, the small pH range for methane formation, and the algal photosynthesis process shifted to the right.

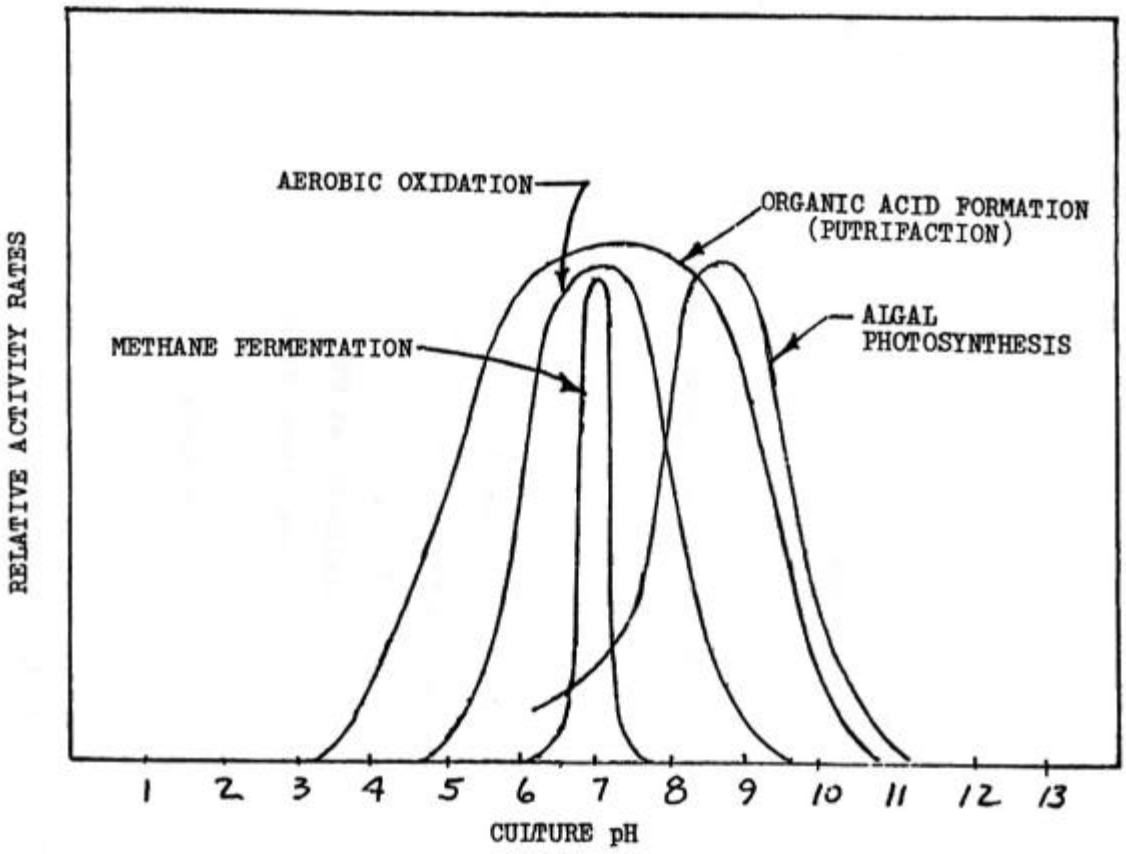

Figure 5.4 pH Activity Curves for Major Microbial Processes in Ponds (Oswald, 1994)

In addition, Figure 5.5 shows the average number of organism per 100 mL versus the raw sewage influent and different ponds. As indicated, there is generally a log difference between each step.

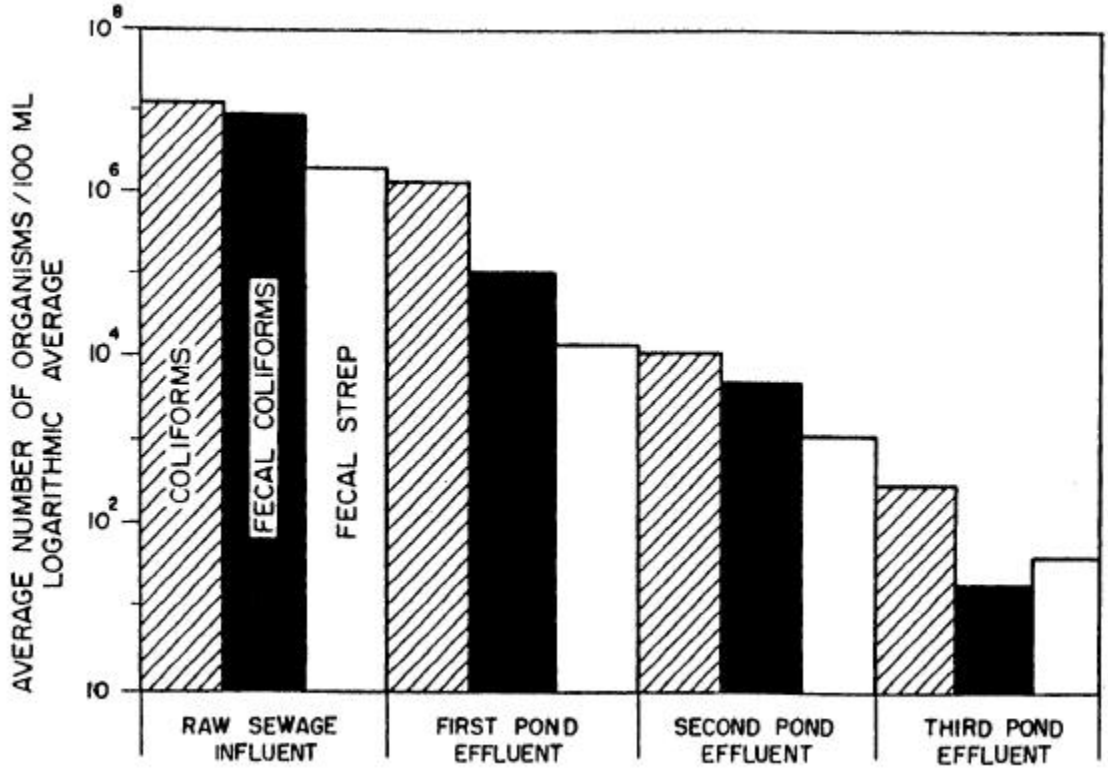

Figure 5.5 The Influence of Series Ponds on Decrease in Average Number of Sewage Bacteria (Oswald, 1994)

Oswald's work is relevant to the discussion of swine waste treatment processes primarily because of the design of the Advanced Facultative Pond in series with the High Rate Pond. These two ponds, working together, are similar in concept to the upflow anaerobic/aerobic system discussed herein. As stated earlier, the AFP is designed to avoid intrusion of dissolved oxygen. Thus, Oswald recommends fermentation pits to insure anaerobic conditions. In the upflow anaerobic/aerobic system, the intent, in part, was to determine whether anaerobic conditions could adequately be met without the pits. In addition, in lieu of an aerobic pond in series with the anaerobic pond, it was the intent, in part, to determine if oxygen could be introduced in a non-turbulent way to the pond in

the upper zone to provide for oxygenation of odorous compounds and assist to some degree in organic loading reductions.

Microbial characterization of a swine lagoon

Researchers Chikh, Pourquie, Kaiser, and Davila (1997) characterized the bacterial flora of a swine lagoon pilot plant. Rationale for the research included odor issues and the concern of field disposal that they claim may contribute to pollution and eutrophication of surface and subsurface waters. This work was performed on a compartmented aerated lagoon system in which the nutrients in the swine manure are first converted into algal biomass and then into zooplankton intended for fish feeding.

The following list shows the type of bacteria analyzed and the methods used to determine the relative magnitude of each:

- total eutrophic flora were analyzed using both anaerobic and aerobic total plate counts (3 days at 30^0C),
- total oligotrophic flora (10 days at 30^0C),
- total coliforms, fecal coliforms, fecal streptococci, and spores of sulfite-reducing clostridia (most probable number),and
- nitrifying and denitrifying flora (most probable number).

The results of this analysis are shown in Table 5.3 and Table 5.4. Each compartment in the research displayed a specific flora, different from the flora in the manure, and consisting of a complex assembly of Gram-negative and Gram-positive ubiquitous species. Of the total number of species identified, 62% were of the *Bacillus*, *Pseudomonas*, and *Aeromonas* genera. The researchers contend that the diverse makeup of the flora is in response to special environmental conditions that prevail in the ponds.

These conditions include variations in pH and dissolved oxygen and low but changing concentrations in available organic matter.

Table 5.3 Enumeration of the Total Flora at Different Sampling Dates Under Eutrophic, Oligotrophic, and Anaerobic Conditions (Chikh, 1997)

Enumeration (10^5 CFU/mL)

Sample Origin	19 May 1995			4 July 1995			28 July 1995		
	Eutrophic	Oligotrophic	Anaerobic	Eutrophic	Oligotrophic	Anaerobic	Eutrophic	Oligotrophi	Anaerobic
Manure	15	nd	0.23	102	nd	1.29	265	nd	0.94
Algal ponds	4.1	165	0.13	4.21	24.2	0.39	0.228	69.5	0.07
Daphnid ponds	0.56	2.67	0.012	0.8	5.9	0.6	0.48	7.2	0.29
Fish pond	0.15	10.6	0.01	0.15	5.3	0.11	0.41	5	0.16

Table 5.4 Enumeration of the Sanitary Flora and Nitrifying Flora (Chikh, 1997)

Enumeration of the sanitary flora

Flora	19 May 1995				4 July 1995			
	Manure	Algal ponds	Daphnid ponds	Fish pond	Manure	Algal ponds	Daphnid ponds	Fish pond
Total coliforms*	45	15	0.2	0.15	0.45	0.009	0.004	0
Fecal coliforms*	0.2	0.2	0.045	0.005	0	0	0	0
Fecal streptococci*	9.5	0.75	0.007	0.015	4.5	0.095	0.007	0
Clostridia*	150	2.5	0.15	0.025	200	3	0.2	0.025
Aeromonas**	6.5	10.4	5	2	11	190	29	11
Listeria**	0	0	0	0	0	0	0	0
Salmonella**	0	0	0	0	0	0	0	0

*Results are expressed in 10^2 MPN
**Results are expressed in 10^2 CFU/mL

Enumeration of the nitrifying flora at different sampling date.

Sampling date	Temperature (^{0}C)	Enumeration (MPN)		
		Algal ponds	Daphnid ponds	Fish pond
15 May 1995	12.2	0.048×10^4	0.068×10^4	0.003×10^4
4 July 1995	19	408×10^4	7×10^4	4×10^4
28 July 1995	21.5	1.55×10^4	1.9×10^4	0.0509×10^4

Pathogenic microbiology in swine lagoons

Additional swine microbial flora analyses have been performed by others as well. Hill and Sobsey (1998) examined the bacterial, viral, and parasitic pathogens found in swine wastes. By collecting samples from a North Carolina swine nursery, they were able to determine the mean concentrations of bacterial indicators such as fecal coliforms, *E. coli*, enterococci, and *C. perfringens* spores. They compared their results and others, to other types of treatment systems. Table 5.5 shows the mean microbial indicator concentrations for alternative treatment systems, and Table 5.6 shows the reductions in mean indicator concentrations for alternative treatment systems.

Table 5.5 Log_{10} Mean Microbial Indicator Concentrations for Alternative Treatment Systems (Hill, 1998)

Sample type	**Indicator type**						
	Faecal Coliforms	***E coli***	**Entero-cocci**	**Total *C perf***	***C perf* spores**	**Somatic phage**	**F+ phage**
Lagoon influent	7.7	7.5	7.4	4.2	4.4	7.4	4.8
Lagoon effluent	5.5	5.4	5.5	4.4	4.2	5.1	3.7
Wetland cell 1 influent	5.3	5.2	5.2	4.1	-	5.0	3.6
Wetland cell 1 effluent	4.4	4.1	4.1	2.9	-	3.8	2.4
Wetland cell 2 effluent	3.9	3.5	3.5	2.6	-	2.5	1.3
Media filter effluent	5.3	4.8	4.8	4.0	-	4.2	2.5
Overland flow effluent	5.1	5.0	5.0	4.2	-	4.7	3.1

Table 5.6 Log_{10} Reductions in Mean Indicator Concentrations for Alternative Treatment Systems (Hill, 1998)

Treatment system	**Indicator type**						
	Faecal Coliforms	***E coli***	**Entero-cocci**	**Total *C perf***	***C perf* spores**	**Somatic phage**	**F+ phage**
Anaerobic lagoon	2.2	2.1	1.9	(0.2)	0.2	2.3	1.1
Constructed wetlands	1.4	1.7	1.1	1.5	-	2.5	2.3
Media filter	0.2	0.6	0.4	0.4	-	0.9	1.2
Overland flow	0.4	0.4	0.1	0.2	-	0.4	0.6
Lagoon + wetlands	3.6	3.8	3.0	1.3	-	4.8	3.4
Lagoon + media filter	2.4	2.7	2.3	0.2	-	3.2	2.3
Lagoon + overland flow	2.6	2.5	2.0	0	-	2.7	1.7

With specific regard to the anaerobic lagoon, fecal coliform in the raw swine wastewater was 4.6×10^7 CFU/100 mL and *E. coli* was 2.9×10^7 CFU/100 mL. The effluent from the anaerobic lagoon had a mean fecal coliform concentration of 3.3×10^5 CFU/100 mL. This concentration is well above state regulations and federal guidelines for maximum allowable fecal coliform levels in municipal wastewaters applied to land in the U.S. The concern, therefore, is in regions having highly porous subsurface matrices, and the possibility of pathogenic groundwater contamination.

CHAPTER VI

PREVIOUS SWINE ODOR REDUCTION RESEARCH

The previous discussion on ponds and characteristics of the microflora related specifically to swine wastewaters provides a basis for understanding the physiology of swine wastewater systems. Additionally, other researchers have investigated methods to address the disadvantages of these pond systems. This research has resulted in the issuance of several patents and publications. The following is a review of several key research findings.

Treatment system - patent

Kolber (1999) recommends, and filed for patent, a treatment system for handling noxious odors and water pollution associated with raising of hogs, cattle, or poultry under confined conditions. His design, as shown in Figure 6.1, replaces the conventional waste lagoon and spreading fields with as wastewater treatment plant. Kolber cites several reasons for changing the current methodology of handling of swine waste. These include the undesirable odor component, the leaching possibilities of lagoon systems, the potential overflow of waste materials into surface runoff tributaries, the possibility of *Pfresteria* outbreaks.

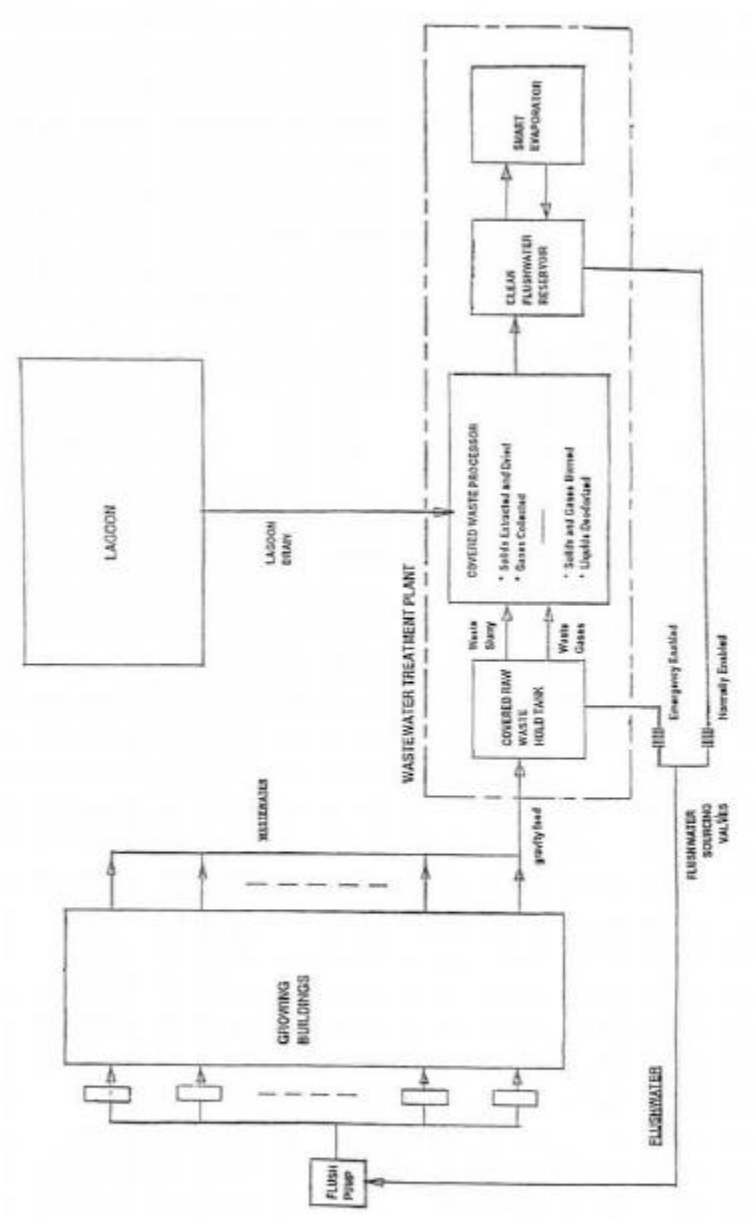

Figure 6.1 Treatment of Waste by Farm Animals Raised Under Confined Conditions

In brief, the method begins by establishing a wastewater stream of flushwater and waste from the confined pen area. The manure is separated into a wet manure portion and a liquid portion. The wet portion is dewatered and burned. The liquid portion is cleansed and recirculated to the confined animal growing area as flushwater. The waste gases produced during the separating and dewatering step may be collected and burned. In handling the treatment of the waste in this fashion, Kolber contends the invention does not produce ammonia and methane gases, or other deleterious by-products.

Odor control system - patent

Mason and Dechant (1999) suggests that wastewater lagoons be treated with compounds of chlorine and oxygen to react with odorous sulfides and mercaptans. This invention, as shown in Figure 6.2, applies a fine spray of an aqueous solution of a chlorine oxygen compound (HOCl, $NaClO_2$, or NaOCl) over the lagoon. The chlorine oxygen compounds react with the sulfides in the vapor zone above the lagoon, converting the odorous compounds into non-odorous compounds. The following equations indicate the resultant reactions:

A) Chlorite

$ClO_2^- + H_2S \longrightarrow Cl^- + 2S^0 + 2H_2O$ Eq. 6.1

$2ClO_2^- + H_2S \longrightarrow H_2SO_4^- + 2Cl^-$ Eq. 6.2

B) Hypochlorite

$OCl^- + H_2S \longrightarrow S^0 + Cl^- + H_2O$ Eq. 6.3

$4OCl^- + H_2S \longrightarrow SO_4^{=} + 2H^+ + 4Cl^-$ Eq. 6.4

C) Cl_2 gas in water to form hypochlorous acid ($Cl_2 + H_2O \longrightarrow HCl + HOCl$)

$HOCl + H_2S \longrightarrow S^0 + Cl^- + H_2O$ Eq. 6.5

$$4HOCl + H_2S \longrightarrow SO_4^{=} + 4Cl^{-} + 5H^{+} \qquad \text{Eq. 6.6}$$

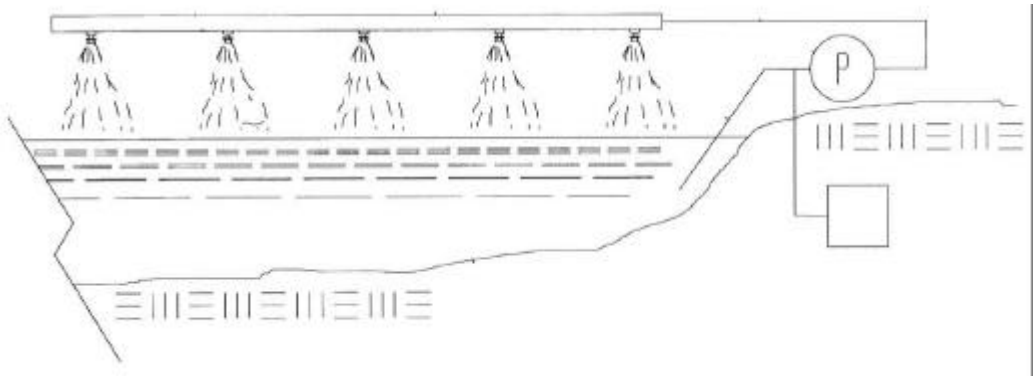

Figure 6.2 Treatment of Odorous Compounds from Wastewater Lagoons by Spraying of Chlorine Oxygen Compounds (Mason, 1997)

The patent application states that the most common source of odors in wastewaters are reduced sulfur compounds such as H_2S and mercaptans which result from the anaerobic decomposition of biodegradable organic matter in the presence of sulfates. In most aerobic digestive systems, odor is generally not a problem. Thus, this invention is directed primarily at anaerobic digestive systems.

Sulfide precipitation - patent

Green and Dowell (1995) suggest an invention to reduce or eliminate odor problems from lagoons systems caused by hydrogen sulfide by the addition of iron compounds such as ferrous or ferric chloride. It is believed that the iron compounds reduce the amount to hydrogen sulfide by precipitating sulfide ions as iron sulfides. The inventors contend that the quantity of iron compound required to reduce the proportion of hydrogen sulfide to an acceptable level is significantly more than the quantity which could be expected by calculation based on the quantity of sulfide present in the sewage.

According to the inventors, the procedure involves adding to the sewage a water-soluble compound of iron at a concentration between 0.2 and 4.0 mg/L calculated as weight of Fe/L of sewage. This compound of iron would be added prior to any primary settlement or clarification at a treatment plant or lagoon. In general, however, it was determined that a concentration between 1.0 and 2.0 mg Fe/L of sewage was preferred.

The iron compounds suggested for this process include ferrous chloride, ferric chloride, ferrous nitrate, ferrous sulfate, ferric sulfate, and ferrous acetate.

Swine odor reduction patent summary

Each of the patent processes previously described has advantages and disadvantages associated with them. These characteristics are summarized in the Table 6.1. As indicated there are common disadvantages in first cost and operating cost associated with each patent process. Each disadvantage shown is extremely important to the CAFO owner as each will impact the profitability of the operation.

Stratified facultative lagoon

Schulz and Barnes (1990) performed experiments on a stratified facultative lagoon utilizing surface aerators of an otherwise anaerobic lagoon to provide a non-odorous cover for the anaerobic contents. The project experiment was performed on two large swine operations, one on the outskirts of Sydney, Australia (the Menagle Piggery) and the other was located in Corowa, New South Wales (the Corowa Piggery). The research contends that the critical design parameters included lagoon depth, specific energy input, and aeration system design.

Table 6.1 Summary of Odor Reduction Treatment Processes

Process	Advantages	Disadvantages
1. Dewater/Burn/ Recycle/Evaporate	No ammonia No methane No odor No lagoon leaching No lagoon runoff No pathogens	First cost Operating cost Air pollution from burning Air pollution from evaporation Air discharge permitting Availability of fuel source Variation in fuel price System complexity
2. Chlorine spray	Reduced odors from H_2S and mercaptans	First cost Operating cost Potential chlorine fumes and odors Handling of chlorine materials System complexity
3. Fe compound addition	Reduced H_2S	First cost Operating cost Solids removal Solids disposal Additional material handling Iron injection system

In the case of the Corowa Piggery experiment, the lagoon was 120 meters long, 60 meters wide, and 8 meters deep. The aerator system was comprised of eight 5.9-kilowatt Flygt ejector units as shown in Figure 6.3.

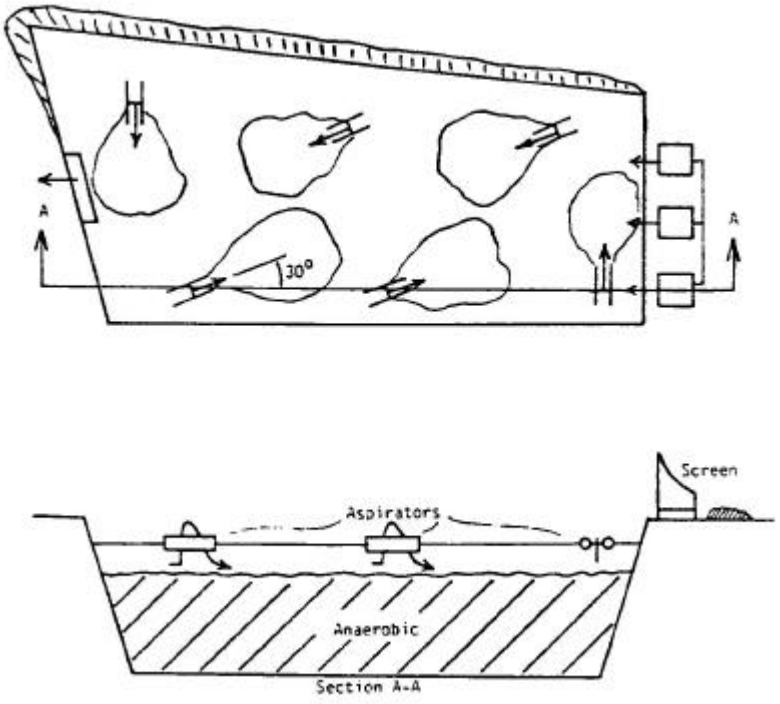

Figure 6.3 Stratified Lagoon Schematic (Schulz, 1990)

In summary, the lagoons were a reliable and effective method for a non-odorous swine waste treatment process. The process removed 75% of the organic materials, and used only one-third of the power required for a fully aerobic system. It was also determined that mean surface redox potentials (E_h) as low as –76 mV are not accompanied by the emission of objectionable odors, despite the absence of dissolved oxygen. The stratified redox potentials are shown in Figure 6.4.

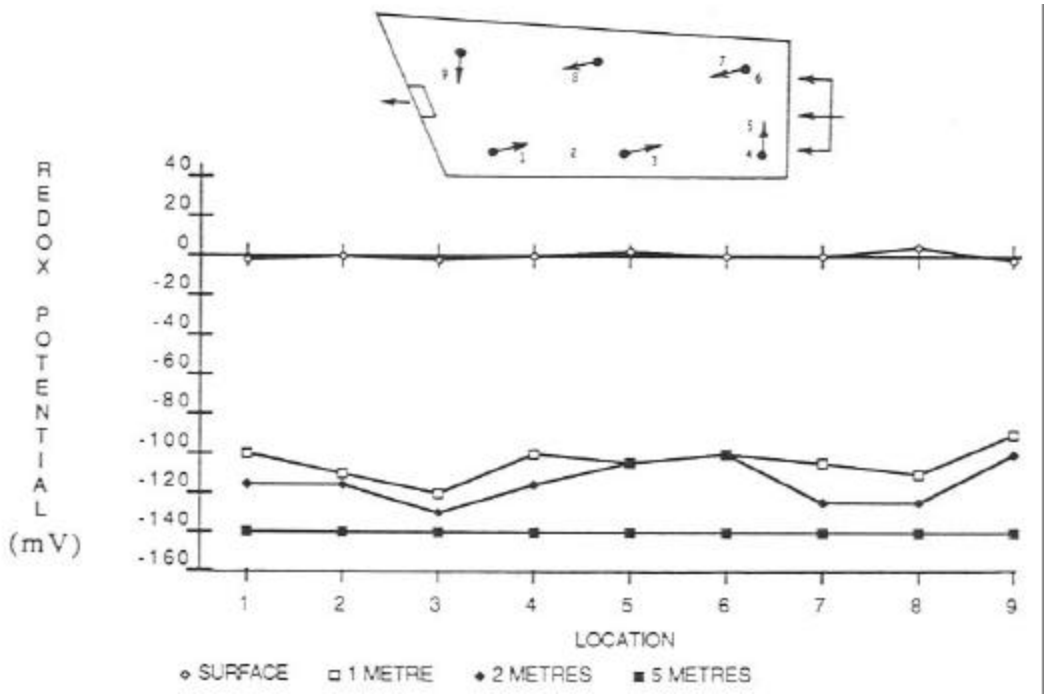

Figure 6.4 Stratified Lagoon – Stratified Profile (Schulz, 1990)

Other lagoon and odor related research

In addition, many others have looked at different aspects of handling swine lagoon and odor issues. Sneath and Williams (1990) examined the importance of wind aeration on controlling odors from a pig slurry after aerobic treatment. They found the effect of wind aeration was potentially far greater than a fourfold increase in the solids residence time of the prior aerobic treatment.

Wong (1990) investigated the possibility of treating pig manure by anaerobic digestion using batch fermentation at 37^0C. In doing so he was able to determine the amount of solids reduction, TOC reduction, total nitrogen, COD reduction, and methane production.

William and Streader (1990) examined methods to predict slurry production on a pig farm. The models developed were based on feed water, slurry relationships, values found in literature, and the digestibility of feed and of water of an actual piggery.

Westerman et al. (1999) examined the impact on odor and treatment using an aerobic fixed-media upflow biofilter for swine manure. The system included a feed tank, two upflow bio-filters, air supply blowers, and a polishing tank. The results of the investigation yielded a significant reduction in odor intensity and irritation as determined by an odor panel, plus adequate removals of COD, SS, nitrogen, and phosphorus.

Westerman and Bicudo (1999) also looked into a treatment design that utilized an aeration and mixing pond for nitrification/denitrification of swine manure. This treatment system consisted of two floating mixers (10 hp each), two floating aerators (30 hp each), a recirculation pump (15 hp), a recycle pump (10 hp), and an overflow to a storage pond. The concept is to convert organic nitrogen and ammonium to nitrate by aeration, and then denitrify the nitrate to nitrogen gas by recycling the nitrate to the front of the system where the waste stream is entering the system. The results of the tests included nitrogen reductions of 65% to 90%, and odor ratings intensity were reduced significantly. The researches noted that the energy costs, however, were high. The system operated 105 horsepower continuously. And, at $0.06 per kWh, this equates to $126 per day. This additional cost decreases the CAFO owners profit by $0.03 per pound by the time the hog is ready for slaughter.

CHAPTER VII

EXPERIMENTAL EQUIPMENT AND PROCEDURES

The reactor

The previous discussion demonstrates the success of reducing odors from wastewater treatment operations wherein odorous vapor compounds are oxidized. The research herein was intended to take advantage of this oxidation effect by modifying the anaerobic lagoon pond system. The concept of the upflow anaerobic/aerobic system is developed by allowing the influent materials to enter into the modified system near the bottom and discharge near the surface, coupled with the addition of oxygen nearer to the surface. The system must be an upflow unit to enhance the possibility of maintaining an anaerobic microbial flora in the bottom and an aerobic microbial flora in the top. In additional, the upflow concept permits the gases created from the microbial activity to be oxidized as they rise to the surface and travel vertically through the aerobic flora.

To accomplish the upflow objective, a 32-ft^3 reactor was constructed. The reactor is shown schematically in Figure 7.1. Figure 7.2 shows the actual reactor. It stood eight feet tall and was equipped with sample ports located every 12 inches. Two thermostat probes and heating elements were inserted into the reactor, one located at 1'-6" from the bottom and the other located at 4'-6" from the bottom. Two air distribution tubes were

located 4’-9” from the reactor bottom. These air distribution tubes were supplied with compressed air through the use of an air compressor and rotometer. The substrate charge was consistently introduced into the bottom of the reactor at the inlet port located 6” from the reactor bottom. The temperature of the reactor was held constant throughout the experiment at 80 degrees F. Air was introduced for most of the experiment at 4 SCFH, which equates to about 0.062 lb O_2/hr.

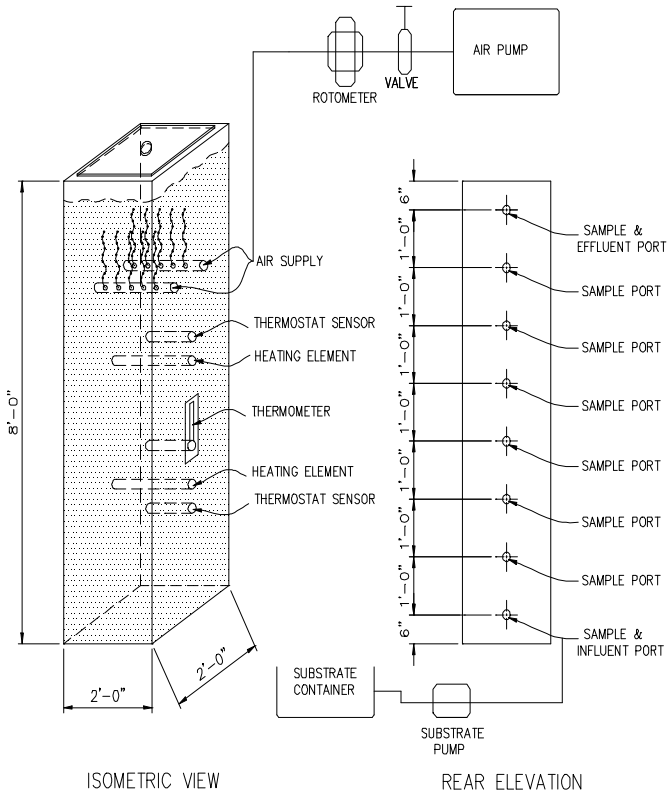

Figure 7.1 Schematic of Upflow Anaerobic/Aerobic Swine Treatment Reactor

Figure 7.2 Upflow Anaerobic/Aerobic Swine Treatment Reactor

Working in with a local hog-finishing farm, 200 gallons of anaerobic sludge was obtained from the CAFO's anaerobic lagoon. The sludge was pumped from the lagoon bottom at a depth of approximately ten feet to insure the sludge was anaerobic. The sludge was then introduced into the reactor. Next, to simulate the operation of the anaerobic lagoon, substrate material was taken twice weekly from the CAFO pit area and introduced to the reactor. This procedure was performed for the following four weeks thus allowing the bacteria to become acclimated to their new environment and substrate loading. A timeline of events is shown in Figure 7.3.

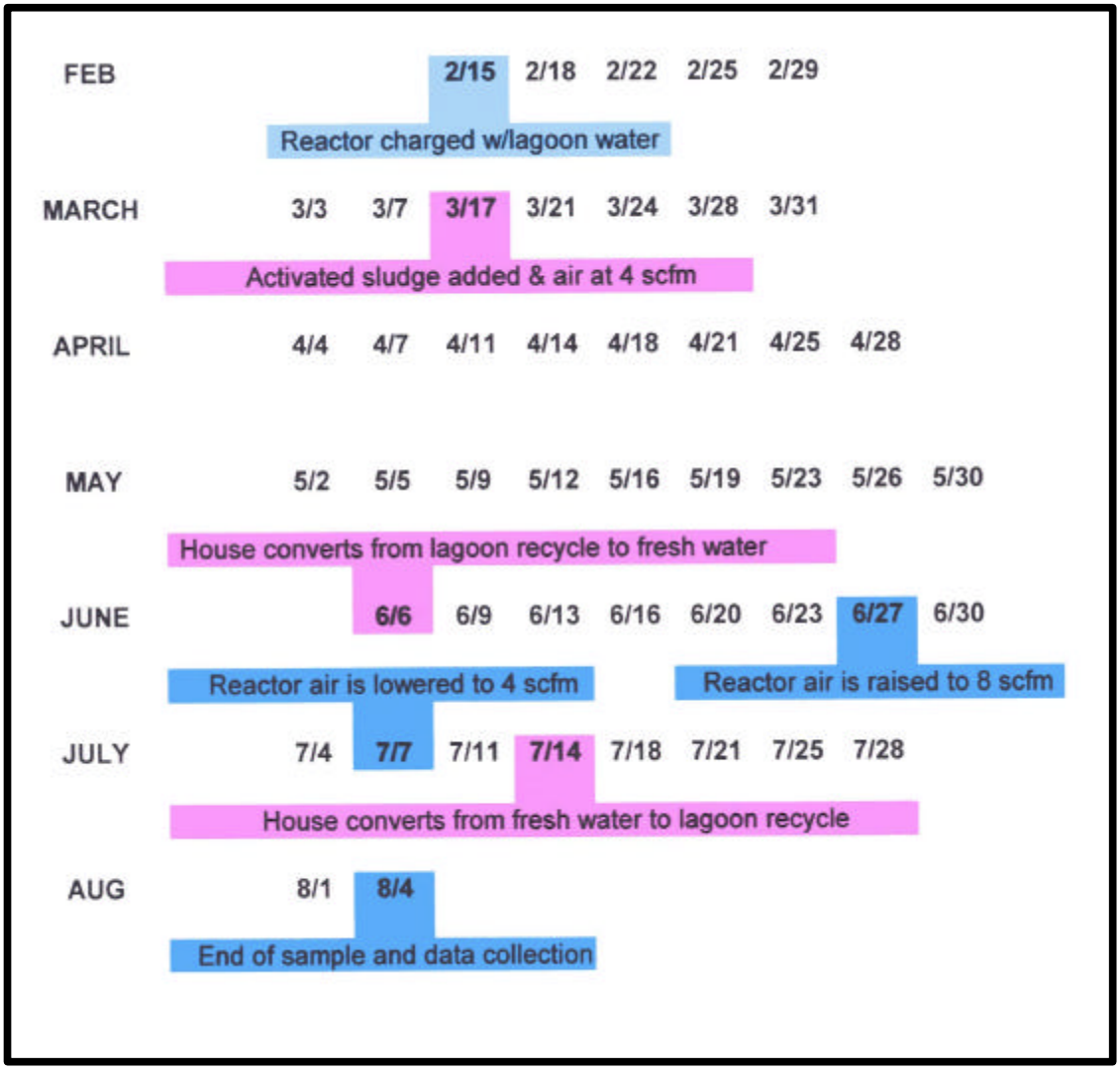

Figure 7.3 Timeline of Pilot Scale Testing of an Upflow Anaerobic/Aerobic Swine Treatment Project

As indicated by the timeline, two gallons of activated sludge obtained from a local municipal wastewater treatment facility was added to the reactor on day 32. The sparging of air at 4 SCFH into the reactor was also initiated at that time. For the next

four months, substrate material taken from the CAFO pit area was introduced twice weekly to the reactor.

The only deviations to the experimental procedures occurred as shown on the timeline. The first deviation occurred when fresh water was used in the house as flushwater instead of recycled lagoon water. This event was an operational change at the CAFO due to lower than desired lagoon levels caused by evaporation. The second deviation was a research parameter change to determine the change in microbial activity brought on by an increase in dissolved oxygen. This change was accomplished by raising the volume of air supplied to the reactor from 4 SCFH to 8 SCFH, and then back again.

The CAFO

The CAFO confinement houses associated with this research is shown in Figure 7.4 and Figure 7.5. This CAFO consisted of eight houses with the capacity of 968 hogs per house. Each house has two rows of hog pens. Altogether there are 44 pens per house, with a capability of 22 hogs in each pen. The hogs lay or stand on a concrete grate system that allows for their urine and feces to pass through the grates and into a concrete holding structure below. This holding structure is the width of the hog pens and runs the entire length of the house. It maintains a recycled lagoon water depth of 12". Thus, the normal operating volume of material in the holding structure of one house is approximately 7,216 ft^3. The urine and feces from the hogs mixes with the recycled lagoon water. Approximately every four days, this holding structure is drained and replenished with 'fresh' lagoon water. The drained contents of the holding structure flow by gravity to the anaerobic lagoon. With the average hog body weight of 150 pounds, 15.0 liters per day of excrement can be expected from a single animal

(http://www.hogwatch.org/enviroimpacts). This excrement consists of both feces and urine with 90% being moisture. Thus, for a house with 968 hogs, 14,520 liters or 3,836 gallons of excrement per day can be expected.

Figure 7.4 CAFO Houses – Front View

Figure 7.5 CAFO Houses – Rear View

The lagoon

Most lagoons handling swine wastewaters from CAFOs are designed in conjunction with the U.S. Department of Agriculture Natural Resources Conservation Service (http://www.ftw.nrcs.usda.gov/tools/awm.hmtl). The NRCS provides a worksheet for determining the size and shape of the lagoon as well as other parameters including pipe sizes and inverts. The NRCS methodology takes into account the following:

- Animal Type
- Number of Animals
- Average Animal Weight
- Manure Volume
- Treatment Period
- Confinement Period
- Daily Wastewater Volume
- Volatile Solids
- Sludge Volume Accumulation Rate
- Sludge Volume Accumulation Period
- Precipitation
- Watershed Area

Table 7.1 shows a completed NRCS worksheet for design of this anaerobic lagoon.

Table 7.1 NRCS Worksheet – Anaerobic Lagoon Design

Decisionmaker _____ Date _____
Site _____

Animal units

1. Animal type *Growers*
2. Animal weight, lbs (W) *150*
3. Number of animals (N) = *7,744*
4. Animal units, AU = W x N/1000 = *1,161*

Manure volume

5. Daily volume of daily manure production per AU, ft^3/AU/day (DVM) = *1.0*
6. Treatment period, days (D) = *180*
7. Total volume of manure production for animal type for treatment period, ft^3 VMD = AU x DVM x D = *208,980*
8. Total manure production for treatment period, ft^3 (TVM) = *208,980*

Wastewater volume

9. Daily wastewater volume per AU, ft^3/AU/day (DWW) = *0*
10. Total wastewater volume for animal description for treatment period, ft^3 WWD = DWW x AU x D = *0*
11. Total wastewater volume for treatment period, ft^3 (TWW) = *0*

Clean water volume

12. Clean water added during treatment period, ft^3 (CW) = *0*

Waste volume

13. Waste volume for treatment period, ft^3 WV = TVM + TWW + CW = *208,980* + *0* + *0* = *208,980*

Manure total solids

14. Daily manure total solids production, lbs/AU/day (MTS) = *6.34*
15. Daily manure total solids production for animal type, lbs/day MTSD = MTS x AU = *7,361*
16. Total manure total solids production lbs/day (TMTS) = *7,361*

Manure volatile solids

17. Daily manure volatile solids production per AU, lbs/AU/day (MVS) = 5.4
18. Daily manure volatile solids production for animal type per day, lbs/day MVSD = AU x MVS = *6,269*
19. Total manure volatile solids production, lbs/day (TMVS) = *6,269*

Wastewater volatile solids

20. Daily wastewater volatile solids production, lbs/1000 gal (DWVS) = *0*
21. Total wastewater volatile solids production for animal type, lbs/day WVSD = (DWVS x DWW x 7.48)/(D x 1,000) = *0*
22. Total wastewater volatile solids production, lbs/day (TWVS) = *0*

Total volatile solids (manure and wastewater)

23. Total daily volatile solids production, lbs/day TVS = TMVS + TWVS = *6,269* + *0* = *6,269*

Minimum treatment volume

24. Selected lagoon VS loading rate, lbs VS/1,000 ft^3 (VSLR) = *6.0*
25. Minimum treatment volume, ft^3 MTV = (TVS x 1,000)/VSLR = (*6,269* x *1,000*)/*6.0* = *1,044,900*

Sludge volume requirement

26. Sludge accumulation ratio, ft^3/lb TS (SAR) = *0.0485*
27. Sludge accumulation period, years (T) = *10*
28. Sludge volume requirement, ft^3 SV = 365 x TMTS x T x SAR = *365* x *7,361* x *10* x *0.0485* = *1,303,080*

Minimum lagoon volume requirement

29. Minimum lagoon volume requirements, ft^3 (MLVR) = MTV + SV + WV = *1,044,900* + *1,303,080* + *208,980* = *2,556,960*

Lagoon sizing

30. Sizing by trial and error $V = (4 \times Z^2 \times d^3)/3 + (Z \times BL \times d^2) + (Z \times BW \times d^2) + (BW \times BL \times d)$

V must be equal to or greater than MLVR = *2,556,960* ft^3

Slide slope ratio, (Z) = *2.0*

Trial no.	Bottom width ft (BW)	Bottom length ft (BL)	Depth ft (d)	Volume ft^3 (V)
1	320	650	10	2,279,333
2	320	650	12	2,784,576
3	320	650	11	2,529,839
4	320	650	11.5	2,656,676

Depth Adjustment

31. Depth adjustment

Depth, ft (d)	11.5
Add depth of precipitation less evaporation on lagoon surface	+ 0.6
Add depth of 25-year, 24-hour storm	+ 0.5
Add for freeboard (1.0 foot minimum)	+ 1.0
Final depth	13.6

32. Compute total volume using final depth, ft^3 *3,201,038*

As seen from this worksheet the total NRCS lagoon volume requirement was 3,201,038 ft^3, with the minimum NRCS treatment volume being 1,044,900 ft^3. Thus the acres required for this 13.6 feet deep anaerobic lagoon is 5.4 acres. The lagoon built for this facility is shown in Figure 7.6.

By comparison, conventional lagoon design would have resulted in a lagoon smaller in size. Table 7.2 shows a summary of conventional design criteria for wastewater stabilization ponds (Ramalho, 1983).

Table 7.2 Summary of Design Criteria for Wastewater Stabilization Ponds

Criteria	Ponds		
	Aerobic	Facultative	Anaerobic
Depth (ft)	0.5-1.5	3-8	8-15
Detention time (day)	2-6	7-50	5-50
Loading			
lb BOD_5/acre day	100-200	200-500	250-4000
BOD removal (%)	80-95	70-95	50-80
Algae concentration (mg/L)	100	10-50	-

From Table 7.2 an anaerobic lagoon can handle as little as 250 lbs BOD_5/acre/day and as much as 4000 lbs BOD_5/acre/day. Based on average influent BOD_5 loading of 350 mg/L at a volume of 57,728 ft^3 per 3.5 days, yields a BOD_5 loading for this CAFO of 360 lbs BOD_5/day. And, assuming a worst-case design scenario of 250 lbs BOD_5/acre day, results in a 15 feet deep, 1.44 acre anaerobic lagoon.

Reactor size and substrate amounts

To obtain the proper lagoon/substrate ratio for the pilot reactor, the following analysis was performed:

From the NRCS form, the total treatment lagoon volume is 3,201,038 ft^3 or 23,943,764 gallons, at a lagoon depth of 13.6 feet. This would result in a lagoon area of 5.4 acres. And, based on the size of the houses:

20.5 ft wide * 176 ft long * 1ft deep * 2 isle/house * 8 houses

= 57,728 ft^3 or 431,805 gallons

Thus, total flush cycle volume = 57,728 ft^3 or 431,805 gallons

Therefore, the lagoon to substrate ratio is:

23,943,764/431,805 = 55.5 to 1

And, the working reactor volume was:

2ft * 2ft * 7.5ft tall = 30ft^3 or 225 gallons

Thus, the minimum substrate required was 225/55.5 = 4.0 gallons.

This figure represents the NRCS minimum biweekly substrate amount to be used on the pilot reactor. However, in examining conventional biological loading design parameters versus NRCS design parameters for the minimum treatment volumes required, the following can be seen:

Acres required – conventional design parameters = 1.44

Acres required – NRCS design parameters = 5.4

Thus, the amount of substrate to be added biweekly to the pilot reactor could have ranged from 4 gallons (NRCS design) to 15 gallons (ie., 4 x 5.4/1.44, conventional design). Therefore, to adequately test the capability of the pilot reactor and in effort to minimize the treatment volume required, it was decided to double the volume currently entering into the NRCS designed lagoon. Thus, 8.0 gallons of substrate was used biweekly to load the pilot reactor.

Figure 7.6 CAFO Lagoon

This CAFO consisted of eight houses. Normal pig loading and unloading procedures resulted in the pigs in each house to be approximately one week older than the pigs in the adjacent house. Thus, to maintain consistency in the samples, the substrate was collected from two different houses with 4 gallons coming from house #3, and 4 gallons coming from house #6. This method of collection provided a consistent concentration of substrate throughout the collection period. The substrate was collected in an 8-gallon container with the assistance of a drum-pump. The material was then taken to the reactor and pumped into it through the bottom inlet port located 6 inches from the bottom of the reactor.

Sample collection procedure

Throughout the project, each time substrate materials were collected from the CAFO house, a representative sample was analyzed. In addition, prior to introducing the substrate into the reactor, samples were taken from the reactor at the bottom sample port (i.e., 6 inches from the bottom) and from the reactor at the top sample port (i.e., 6 inches below the liquid surface). The volume of substrate added biweekly matched the amount of effluent drained from the reactor at the top sample port.

The three samples (substrate, reactor bottom, and reactor top) were then analyzed immediately or placed into a freezer at 3^0C, and sample analysis run within 48 hours.

Sample analysis

All degradation activity and resulting odors are the result of the microbial flora. It is important, therefore, to study the physiology of the microbial population and understand causation of specific observances. Therefore, to adequately establish the success or failure of this project, the parameters necessary to be analyzed were of extreme importance. The project had to demonstrate its effectiveness from two perspectives: biological organic loading reductions and microbiotic physiology related to offensive off-gas production.

From a microbiotic perspective, the parameters analyzed included carbohydrate utilizers that produce gas and acid, denitrifiers, sulfate reducers, anaerobic total plate counts, aerobic total plate counts, and hydrogen sulfide producers. In addition, related parameters of interest included ammonia-nitrogen, nitrate, sulfide, and sulfate. The parameters analyzed from a biological organic loading perspective included pH, dissolved oxygen, BOD_5, total suspended solids, and volatile suspended solids.

The following discussion describes the methods used to perform the sample analyze.

Dilutions

For every sample analyzed serial dilutions were made. Using 100-mL bottles, 1 to 10 dilutions were prepared for each sample using 45 mL of distilled, autoclaved water, and 5 mL of sample or sample dilution.

Total plate counts - aerobic

Using Bacto Plate Count Agar, 10 microliters of sample were spread on a 60 mm diameter by 15 mm deep Petri dish. Duplicates were made for each dilution sample. The plates were placed in an incubator at 30^0C for 48 hours, after which time the number of colony-forming units were counted and recorded. Only plates containing between 30 and 300 colonies were considered countable. The cell density was then determined based on the following formula (Leboffe and Pierce, 1996):

Cell density = # CFU/(Volume plated x dilution factor)

The following figure shows a Petri dish with the results of this analysis.

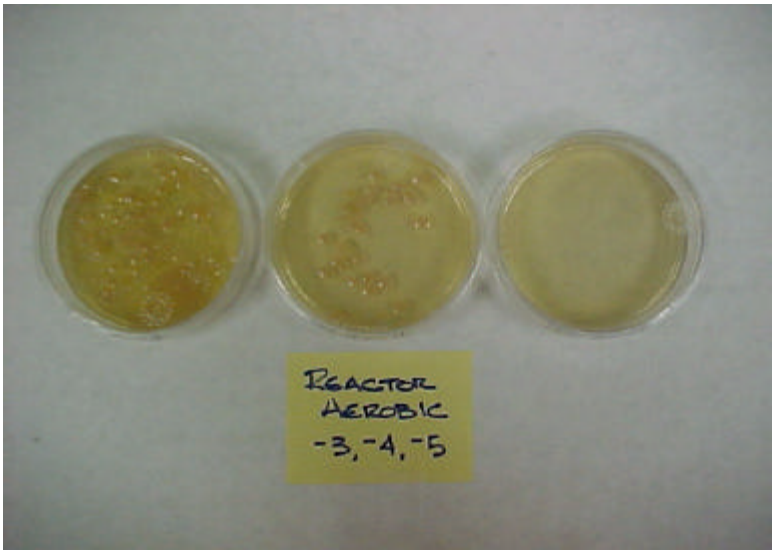

Figure 7.7 Total plate count - aerobic

Total plate counts - anaerobic

Using Bacto Plate Count Agar, 10 microliters of sample were spread on a 60 mm diameter by 15 mm deep Petri dish. Duplicates were made for each dilution sample. The plates were placed into an anaerobic jar and the oxygen was removed by the use of BBL GasPaks^{TM} and catalysts. The anaerobic jar was placed in an incubator at 30^0C for 72 hours, after which time the number of colony-forming units were counted and recorded. Only plates containing between 30 and 300 colonies were considered countable. The cell density was then determined based on the following formula (Leboffe and Pierce, 1996):

Cell density = # CFU/(Volume plated x dilution factor)

The following figure shows an anaerobic jar apparatus.

Figure 7.8 Anaerobic jar

Hydrogen-sulfide producers

Using Bacto Peptone Iron Agar, 10 microliters of sample were spread on a 60 mm diameter by 15 mm deep Petri dish. Duplicates were made for each dilution sample. The plates were placed into an anaerobic jar and the oxygen was removed by the use of BBL GasPaks^{TM} and catalysts. The anaerobic jar was placed in an incubator at 30^{0}C for 72 hours, after which time the number of colony-forming units were counted and recorded. Only plates containing between 30 and 300 colonies were considered countable. The cell density was then determined based on the following formula (Leboffe and Pierce, 1996):

Cell density = # CFU/(Volume plated x dilution factor)

The following figure shows three hydrogen-sulfide producing dilution plates.

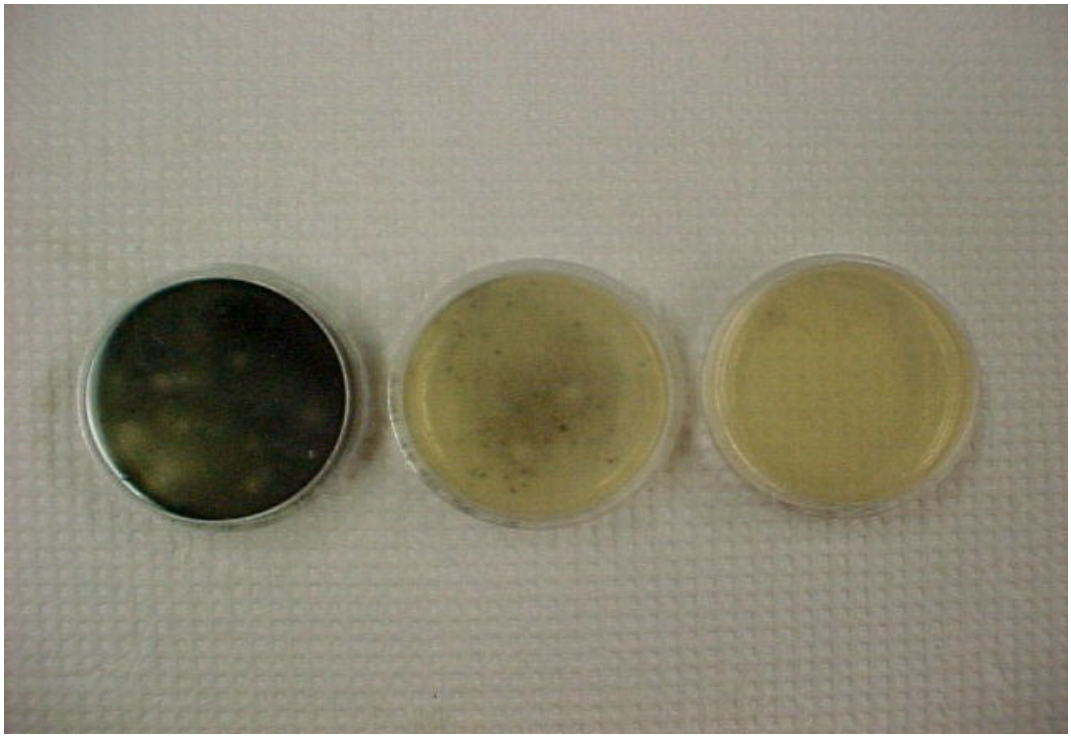

Figure 7.9 Hydrogen-sulfide producers

Carbohydrate utilizers

This parameter was quantitatively analyzed using the MPN (Most Probable Number) method. Using a Bacto Glucose Broth with a phenol red pH indicator, in test tubes fitted with Duram fermentation tubes were prepared and 9 mL of broth were placed in each, sterilized by autoclaving, and stored until time of use. The tubes were then arranged into a 3-3-3 configuration for MPN determinations. Into each tube, 1 mL of sample or sample dilution was added. The tubes were placed in an incubator for 48 hours at 30^0C, after which time the tubes were examined for color change (yellow to red, indicating gas production) and for gas in the Duram tubes. These tubes were counted and the most probable number of microorganisms obtained from a statistical table.

The following figure shows three tubes of glucose broth after incubation. The tube on the left is negative for acid and gas. The middle tube is positive for acid, negative for gas. And the tube on the right is positive for both acid and gas.

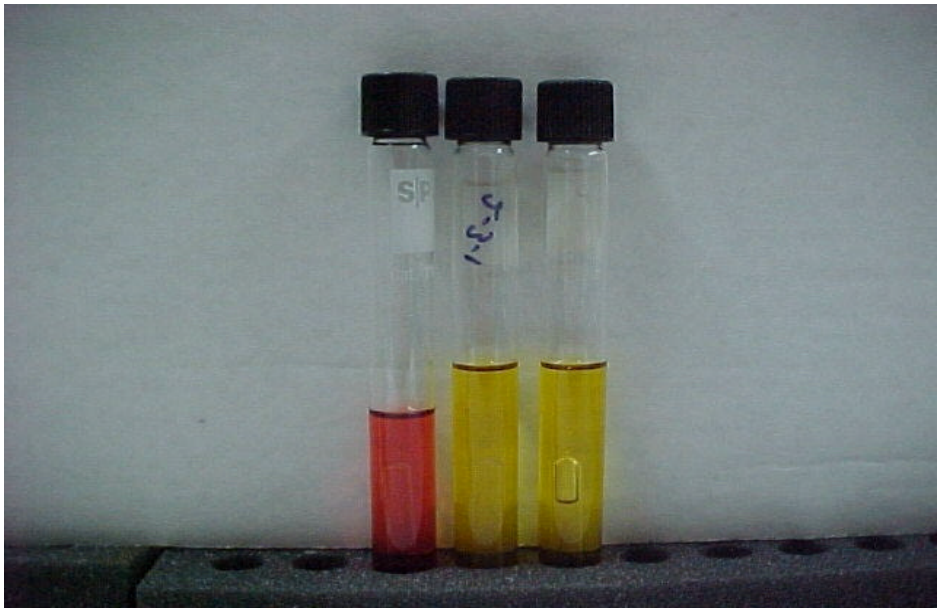

Figure 7.10 Carbohydrate utilizers

Denitrifiers

Using a Bacto Nitrate Broth, test tubes with Duram fermentation tubes were prepared and 9 mL of broth were placed in each, autoclaved, and stored until time of use. The tubes were then arranged into a 3-3-3 configuration for MPN determinations. Into each tube, 1 mL of sample or sample dilution was inserted. The tubes were placed in an incubator for 48 hours at 30^0C, after which time the tubes were examined for gas in the Duram tubes. The results were recorded and the most probable number of denitrifiers determined using a statistical table.

The following figure shows three tubes of nitrate broth after incubation. The tube on the left is negative for gas. The middle and right tubes are positive for gas.

Figure 7.11 Denitrifiers

Sulfate reducers

Using a Sulfate API Broth, test tubes containing 9 mL of broth were autoclaved and stored until time of use. The tubes were then arranged into a 3-3-3 configuration for MPN determinations. Into each tube, 1 mL of sample or sample dilution was placed. The tubes were placed in an incubator for 48 hours at 30^0C, after which time the tubes were examined for color change (to black). These results were recorded the most probable number of sulfate reducers obtained from a statistical table.

The following figure shows three tubes of sulfate API broths. The tube on the left is negative for sulfate reducers. The middle and right tube are positive for sulfate reducers.

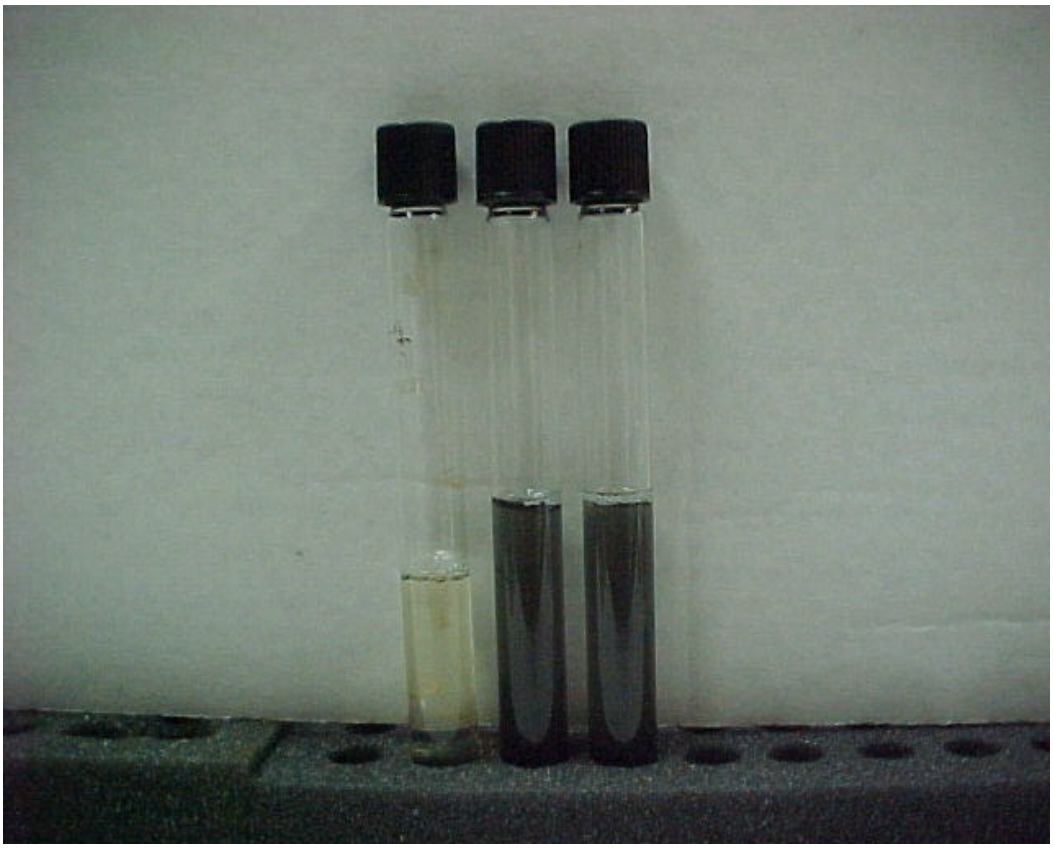

Figure 7.12 Sulfate Reducers

Ammonia-nitrogen

Using the sample dilutions, the direct nesslerization method was used to determine ammonia-nitrogen. A 10 mL sample for up to four sample dilutions was prepared and to each, 8 drops of Reagent #1 (LaMotte #4797-L, 50% potassium sodium tartrate) was added, followed by 1 mL of Reagent #2 (LaMotte #V-4798-L, 15% potassium hydroxide). This solution was thoroughly mixed and allowed to sit until a yellow color appeared in one or more of the vials. In addition, a blank sample was prepared for each of the sample dilutions. With the spectrophotometer set at 420 nm, the appropriate blank sample was used to establish 100% transmittance. The corresponding dilution sample vial was then inserted into the spectrophotometer and % light transmittance was read. And, using a predetermined standard curve for ammonia-nitrogen, the amount of ammonia-nitrogen was determined. This standard curve is shown in Figure 7.13.

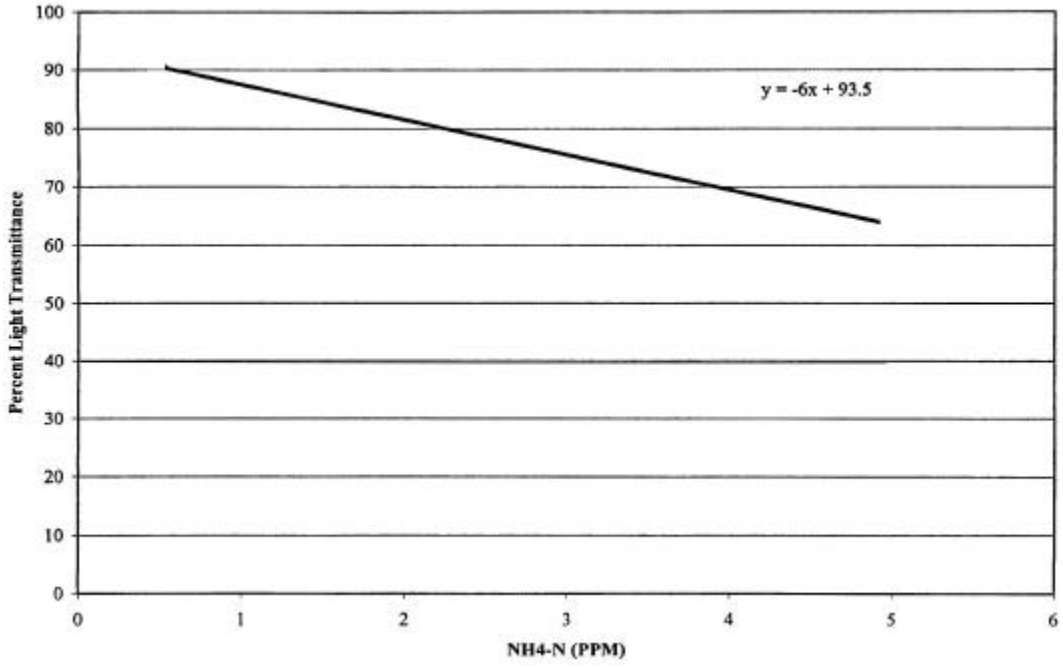

Figure 7.13 Ammonia-nitrogen standard curve

Nitrate-nitrogen

Using the sample dilutions, up to four vials were filled with 5 mL of dilution sample. To these vials, 5 mL of Mixed Acid Reagent (LaMotte #V-6278-L, 17% ammonium chloride, 10% sodium chloride, 4% citric acid, 2% acetic acid, 2% sodium phosphate, 1% copper sulfate) was added and mixed thoroughly. The samples were allowed to sit for a minimum of two minutes. Next, 0.2 grams of Nitrate Reducing Agent (LaMotte #6279D5-G, 10% manganous sulfate, 7% cadmium powder) were added to the tubes and vortexed for one minute. A standard blank of the appropriate sample was inserted into the spectrophotometer set at 220 nm and calibrate accordingly. After 10 minutes, the appropriate sample vial was inserted into the spectrophotometer and absorbance was read as mg/L of nitrate-nitrogen.

Sulfate

Using the sample dilutions, 10 mL samples of up to four sample dilutions were prepared and 0.1 grams of Sulfate Reagent (LaMotte #V-6277-D, 36% barium chloride, 9% citric acid monohydrate) was added to each. The vials were thoroughly mixed and allowed to sit for 5 minutes. In addition, a blank sample was prepared for each of the sample dilutions. The appropriate blank sample was inserted into the spectrophotometer set at 420 nm and calibrated accordingly. The percent light transmittance was read and recorded. And, using a predetermined standard matrix for sulfate, the amount of sulfate present in the sample was determined. This matrix is shown in Table 7.3.

Table 7.3 Sulfate standard matrix

% T	9	8	7	6	5	4	3	2	1	0
90	4.04	4.30	4.56	4.83	5.09	5.36	5.63	5.90	6.17	6.45
80	6.72	7.00	7.28	7.56	7.84	8.12	8.41	8.70	8.99	9.28
70	9.58	9.88	10.18	10.48	10.79	11.10	11.42	11.72	12.04	12.37
60	12.70	13.03	13.36	13.71	14.05	14.40	14.76	15.12	15.49	15.87
50	16.25	16.64	17.04	17.44	17.86	18.28	18.72	19.16	19.62	20.09
40	20.57	21.06	21.57	22.10	22.64	23.21	23.79	24.39	25.02	25.67
30	26.35	27.05	27.79	28.56	29.38	30.23	31.12	32.07	33.07	34.13
20	35.26	36.46	37.74	39.11	40.58	42.17	43.88	45.75	47.77	49.99
10	52.43	55.12	58.10	61.43	65.18	69.41	74.25	79.81	86.29	93.92
0	103.05									

Sulfide

Using the sample dilutions, 10 mL samples of up to four sample dilutions were prepared and 1.0 mL of Sulfide Reagent A (LaMotte #V-4458-L, 64% sulfuric acid, <1% N,N-dimethyl-p-pheylenediamine sulfate) was added to each and the vials were thoroughly mixed. Then, six drops of Sulfide Reagent B (LaMotte #V-4459-L, 25% ferric chloride) was added to each vial, thoroughly mixed, and allowed to sit for one minute. If sulfide ions were present, a blue color developed. For those vials that demonstrated a color change, 2.0 mL of Sulfide Reagent C (LaMotte #4460-L, 40% ammonium phosphate) was added and the samples thoroughly mixed again. In addition, a blank sample was prepared for each of the sample dilutions. The appropriate blank sample was then inserted into the spectrophotometer set at 570 nm and calibrated accordingly. The associated sample vial was then inserted into the spectrophotometer and % light transmittance was read and recorded. And, using a predetermined standard

matrix for sulfide, the amount of sulfide present in the sample was determined. This standard matrix is shown in Table 7.4.

Table 7.4 Sulfide standard matrix

% T	9	8	7	6	5	4	3	2	1	0
90				0.00	0.02	0.03	0.05	0.06	0.08	0.09
80	0.11	0.12	0.14	0.15	0.17	0.19	0.20	0.22	0.23	0.25
70	0.27	0.29	0.30	0.32	0.34	0.36	0.37	0.39	0.41	0.43
60	0.45	0.47	0.49	0.51	0.53	0.55	0.57	0.59	0.61	0.63
50	0.65	0.67	0.70	0.72	0.74	0.76	0.79	0.81	0.84	0.86
40	0.89	0.91	0.94	0.96	0.99	1.02	1.04	1.07	1.10	1.13
30	1.16	1.19	1.22	1.25	1.29	1.32	1.35	1.39	1.43	1.46
20	1.50	1.54	1.58	1.62	1.66	1.70	1.75	1.80	1.84	1.89
10	1.95	2.00	2.06	2.12	2.18	2.24	2.31	2.38	2.46	2.54
0	2.63	2.73	2.83	2.95						

Dissolved oxygen

Using the membrane electrode method, samples were analyzed for dissolved oxygen using a portable YSI model 55/12 FT dissolved oxygen probe. The meter was calibrated weekly by reading against saturated air and dissolved oxygen saturated water samples at known temperatures.

Temperature

The temperature of each sample was taken in the field using an Orion model 230A portable meter. The measurement was taken and recorded in ^{0}C, and, for reactor temperature measurements, the probe readings were compared to the mercury thermometer on the side of the reactor.

Biochemical oxygen demand

Prior to running BOD_5 analysis, dilution water was prepared to provide adequate nutrients for bacterial growth. This dilution water included the preparation of the following reagents: phosphate buffer solution, magnesium sulfate solution, calcium chloride solution, and ferric chloride solution.

Sample dilutions at 20^0C were poured into BOD bottles and 1 mL each of phosphate buffer, $MgSO_4$, $CaCl_2$, and $FeCl_3$ solutions per liter of water was added. No seeding was necessary.

After filling the BOD bottles with the dilution samples, the initial dissolved oxygen measurements were taken and recorded. These measurements were performed using a calibrated laboratory dissolved oxygen meter. The dilution bottles were then sealed and incubated for 5 days at 20^0C. After 5 days of incubation, the final dissolved oxygen was taken using the pre-calibrated dissolved oxygen meter.

The formula for determining the BOD_5 is

$$BOD_5, \text{ mg/L} = (D_1 - D_2)/P \qquad \text{Eq. 7.1}$$

where,

D_1 = dissolved oxygen of diluted sample immediately after preparation, mg/L

D_2 = dissolved oxygen of diluted sample after 5 days incubation at 20^0C, mg/L

P = decimal volumetric fraction of sample used.

This procedure followed the protocols as defined by the Standard Methods 507 Oxygen Demand (Biochemical) (Standard Methods, 1998).

Total suspended solids

The sample material was filtered through a weighed standard glass-fiber filter and the residue retained on the filter was dried to a constant weight at 103^0C to 105^0C. The increase in weight of the filter represented the total suspended solids. The formula for this analysis is,

mg total suspended solids/L = ((A – B) x 1000)/sample volume, mL Eq. 7.2

where, A = weight of filter + dried residue, mg

B = weight of filter, mg

This procedure followed the protocols as defined by the Standard Methods 209 C. Total Suspended Solids Dried at 103^0C – 105^0C (Standard Methods, 1998).

Volatile suspended solids

The residual total suspended solids were ignited to a constant weight at 550^0C. The remaining solids represent the fixed total solids while the weight loss on ignition is the volatile solids. The volatile suspended solids represent an approximation of the solid fraction of the wastewater or reactor water. The formula for this analysis is,

mg volatile solids/L = ((A – B) x 1000)/sample volume, mL Eq. 7.3

mg fixed solids/L = ((B – C) x 1000)/sample volume, mL Eq. 7.4

where, A = weight of residue + dish before ignition, mg,

B = weight of residue + dish or filter after ignition, mg, and

C = weight of dish or filter, mg

This procedure followed the protocols as defined by the Standard Methods 209 D. Fixed and Volatile Solids Ignited at 550^0C (Standard Methods, 1998).

pH

The pH of the samples were analyzed immediately after sample collection. An Orion model 230A portable pH meter was used. The meter was calibrated prior to each set of data collected using pH buffer solutions of 4 and 10.

Statistical analysis

As mentioned, the most probable number method was used on several of the microbial count estimates. Also known as the multiple tube fermentation technique, the MPN technique is based on the principle of dilution to extinction (Standard Methods, 1998). The method consists of the following:

Broths containing the proper carbon and nutrient sources are prepared. In addition to the carbon and nutrient sources, indicators may also be included to provide for positive or negative reactions. These indicators may include iron compounds which cause the solution to turn black or pH indicators which simply cause a change in color of the solution. In addition, for detection of a gas, Duram fermentation tubes are placed into each test tube.

Each tube contains 9 mL of broth solution. Sample dilutions are made and 1 mL from each sample dilution is placed into each of three broth tubes. A minimum of three serial sample dilutions must be used. The tubes are stoppered and allowed to incubate for a minimum of 48 hours at 30^0C.

The results for each dilution are reported as a fraction, with the number of positive tubes over the number of negative tubes. The concentration of total bacteria being analyzed is reported as the 'most probable number' per 100 mL. The MPN is based on the application of the Poisson distribution. Standard MPN tables are used to

determine the MPN index and 95% confidence limits for various combinations of positive and negative results. These numbers reflect serial dilutions of the three 10 mL, three 1 mL, and three 0.1 mL portions. If other dilutions are used, the results shown must be multiplied or divided by the appropriate factor.

To examine the relationship between two variables, the Pearson product moment correlation coefficient was used (McClave and Benson, 1988). This relationship provides a quantitative measure of the strength of the linear relationship between parameters *x* and *y*. This coefficient, denoted by *r*, is dimensionless, and always lies between –1 and +1. The relationship is defined by the following set of equations:

$$SS_{xy} = \sum x_i y_i - \frac{\left(\sum x_i\right)\left(\sum y_i\right)}{n} \qquad \text{Eq. 7.5}$$

$$SS_{xx} = \sum x_i^2 - \frac{\left(\sum x_i\right)^2}{n} \qquad \text{Eq. 7.6}$$

$$SS_{yy} = \sum y_i^2 - \frac{\left(\sum y_i\right)^2}{n} \qquad \text{Eq. 7.7}$$

Then, the coefficient of correlation is

$$r = \frac{SS_{xy}}{\sqrt{SS_{xx} SS_{yy}}} \qquad \text{Eq. 7.8}$$

The closer *r* is to 1 or -1, the stronger the linear relationship between the two variables. Then, if *r* = 1 or *r* = -1, all points fall exactly on the same least square line. Positive values of *r*, imply that *y* increases as *x* increases, and negative values of *r* imply that *y* decreases as *x* increases.

CHAPTER VIII

EXPERIMENTAL RESULTS AND DISCUSSIONS

General observations

From the beginning to the end of the project, the data support a four-stage transformation. Key indicators of this conclusion include dissolved oxygen, pH, hydrogen sulfide producers, sulfate reducers, and temperature. Figures 8.1 through 8.5 show the plots of these parameters over the life of the project, as well as the points of transitions. The reactor was operated lasted a total of 172 days with transition points occurring at day 32, day 131, and day 148. Stage I occurred beginning day 1 and ending on day 32. During this time the pilot reactor was functioning as an anaerobic lagoon, with 8 gallons of substrate being introduced twice weekly. During this stage, the anaerobic bacteria in the pilot reactor were becoming acclimated to their new environment. Stage II began on day 32 and ended on day 131. During this period, the pilot reactor was functioning as an upflow anaerobic/aerobic treatment system. Air, at 4 SCFM, was pumped into the reactor at the location previously described, and the reactor temperature was maintained at no less than 80^0F. Stage III began on day 131 and ended on day 148. During this period, the pilot reactor continued to function as an upflow anaerobic/aerobic treatment system, but the air pumped into the unit was increased from 4 SCFM to 8 SCFM. Stage IV began on day 148 and extended to the end of the project.

During this period, all other parameters and inputs remained constant, but the air pumped into the reactor was reduced back to 4 SCFM.

Figure 8.1 shows the dissolved oxygen levels during each of the four stages. As shown, an anaerobic system existed in Stage I with a dissolved oxygen level of less than 0.25 mg/L. With the addition of the 4 SCFM of air into the upper strata of the pilot reactor, the dissolved oxygen in the upper strata immediately rose to an average of 2.2 mg/L, while the lower strata continued to have a dissolved oxygen content of less than 0.40 mg/L. This situation continued throughout Stage II. During Stage III, with the air-input amount increased to 8 SCFM, the dissolved oxygen levels also increased. The upper strata rose from an average of 2.2 mg/L to a maximum of 3.5 mg/L and lower strata increased from an average of 0.3 mg/L to a maximum of 0.75 mg/L. Finally, by reducing the air input back to 4 SCFM, the dissolved oxygen levels returned to those levels as previously found in Stage II. This period constituted Stage IV.

The pH levels throughout the life of the project are shown in Figure 8.2. As indicated, the pH in the upper strata and the lower strata tracked closely together throughout the project. A Pearson correlation coefficient of 0.98 proves this fact. However, the pH trended upward in Stage II of the project, varying from 7.5 to 8.7. The only exception to the rise in pH occurred at the transition from Stage II to Stage III, where the pH dropped to a minimum of 8.5, but then peaked in Stage IV to 9.2. In Stage IV, with a drop in the dissolved oxygen, the pH also dropped to 8.6.

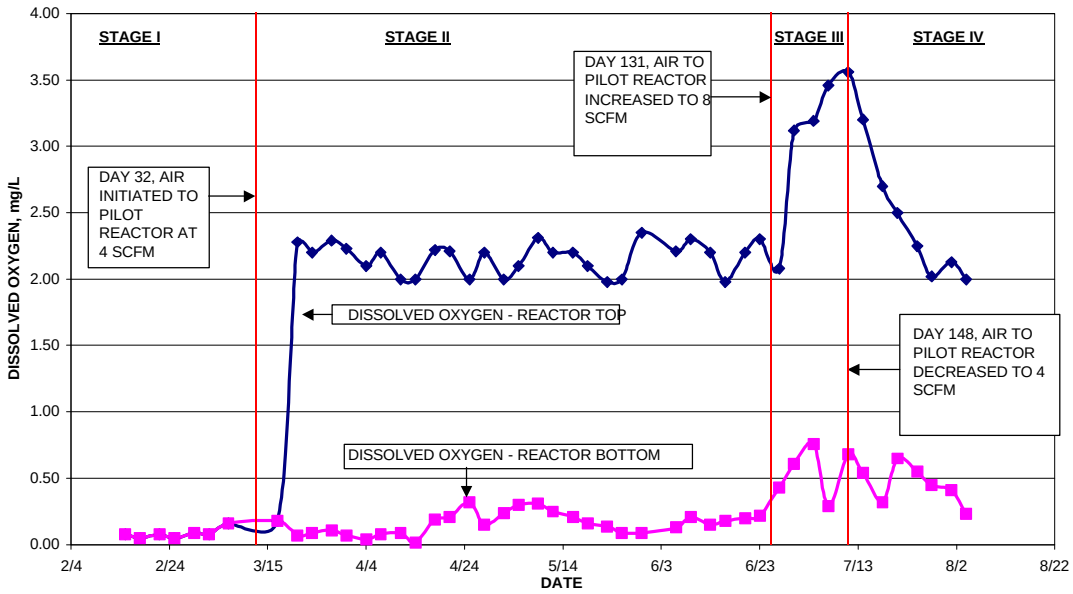

Figure 8.1 Pilot reactor – dissolved oxygen and stage profiles

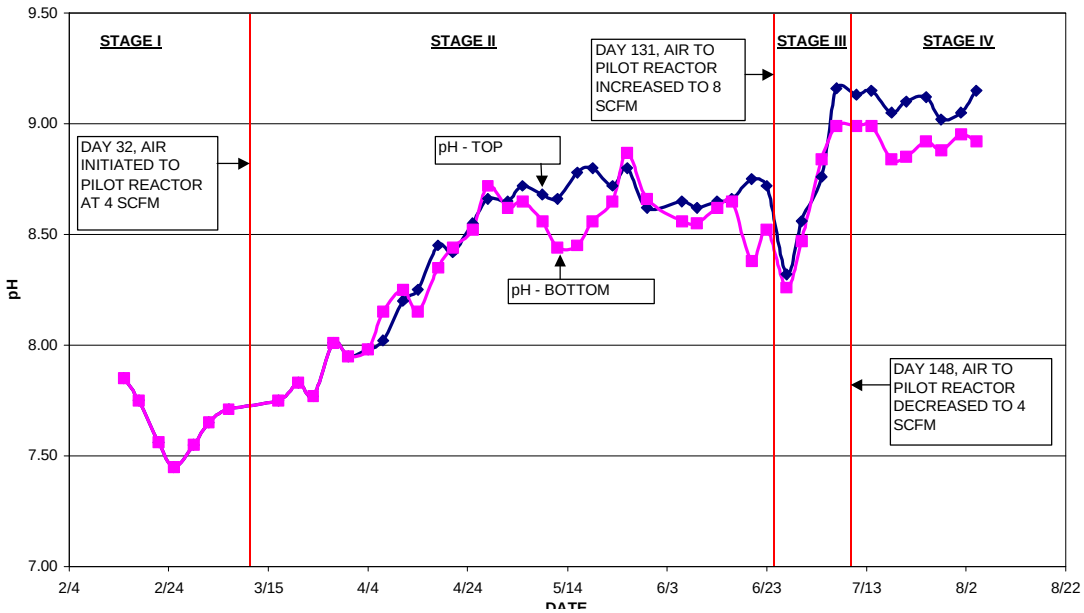

Figure 8.2 Pilot reactor – pH and stage profiles

Hydrogen-sulfide-producing bacteria also contributed to the four-stage conclusion. This fact is indicated in Figure 8.3. In Stage I, hydrogen-sulfide-producing bacteria were prevalent at $1x10^5$ CFU/mL in the upper and lower strata. With the addition of air to the pilot reactor on day 32, the hydrogen-sulfide-producing bacteria in the upper strata dropped, after a lag period of approximately 20 days, to less than 10 CFU/mL. The hydrogen-sulfide-producing bacteria in the lower strata remained consistent at $1x10^5$ CFU/mL. At the beginning of Stage III, the dissolved oxygen level was increased, and there was a slight drop the in hydrogen-sulfide-producing bacteria in the lower strata.

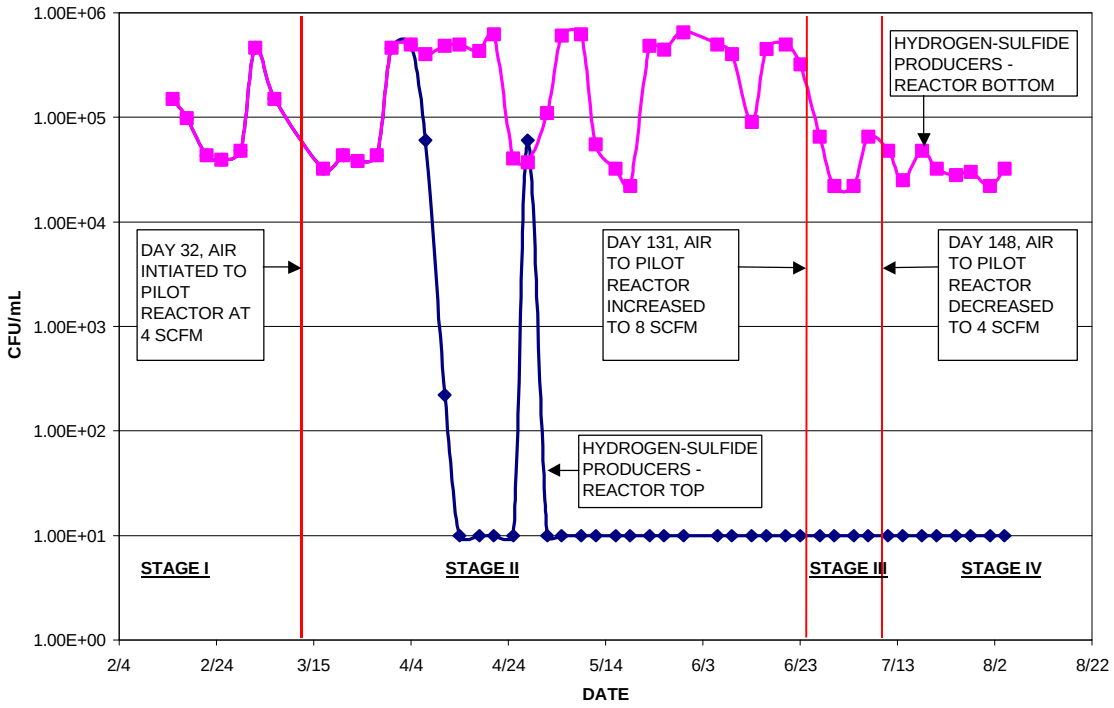

Figure 8.3 Pilot reactor – hydrogen-sulfide-producers and stage profiles

Bacteria found in the pilot reactor that has the ability to reduce sulfate also changed in population during the four transition stages. As indicated in Figure 8.4, sulfate-reducing bacteria were less than 10 (MPN) in both the upper and lower strata during Stage I. After

a period of acclimation, the sulfate-reducing bacteria began to increase in population in the lower strata to a level of approximately $1x10^4$ (MPN). Because of the introduction of air to the pilot reactor prior to the proliferation of sulfate-reducing bacteria in the upper strata, the number of sulfate-reducing bacteria in that stratum never exceeded 10 (MPN). Increasing the air input level from 4 SCFM to 8 SCFM during Stage III had a minimal impact on the sulfate-reducing bacteria in either stratum. In Stage IV, however, by lowering the air input from 8 SCFM back to 4 SCFM, the sulfate-reducing bacteria in the lower strata dropped to less than $1x10^3$ (MPN).

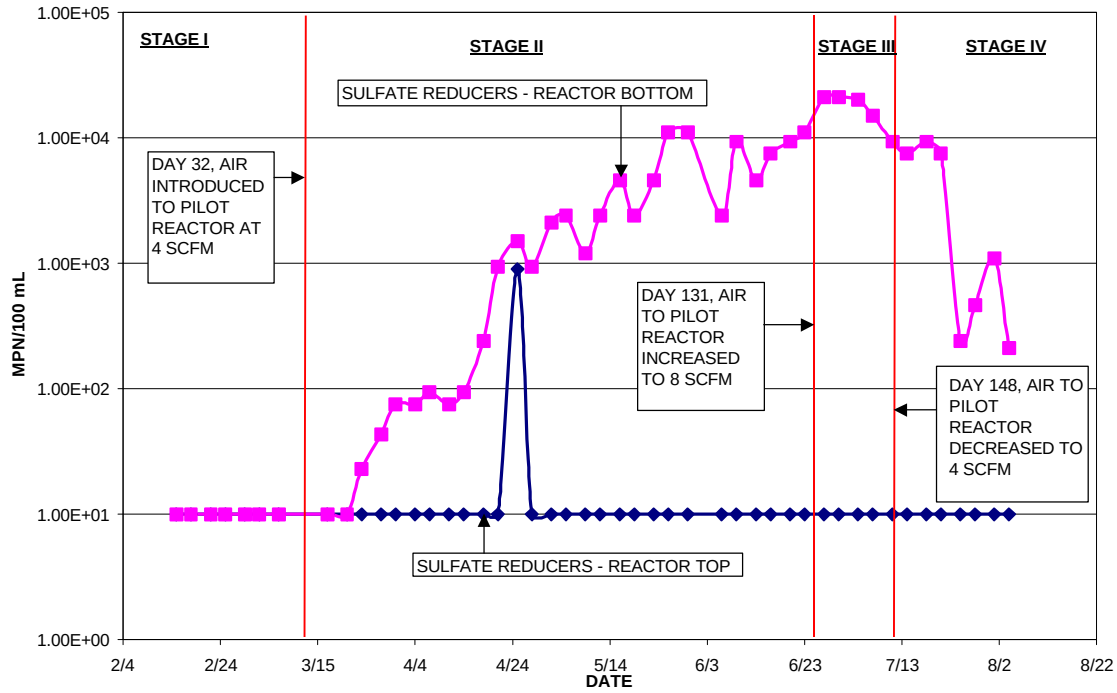

Figure 8.4 Pilot reactor – sulfate reducers and stage profiles

As discussed previously, the reactor was maintained at a temperature level of no less than 80^0F (26.7^0C). Figure 8.5 shows the relationships between the pilot reactor temperature and the substrate temperature as the project moved through each stage. As

indicated, the pilot reactor temperature remained constant at 80^0F through Stage I and Stage II. However, as the project moved into Stage III and warmer spring and summer months, there was for the most part, a consistent increase in the temperature of both the substrate and the pilot reactor. An exception to this observation occurred on day 99 when the CAFO owner initiated fresh-water usage to the house pit areas because of a low lagoon level situation. This process continued until day 130, at which time the CAFO owner resumed the use of recycled lagoon wastewater to the pit areas. The resumption of the lagoon wastewater to the pit areas occurred at approximately the same time as the air input to the pilot reactor was increased from 4 SCFM to 8 SCFM. This fact resulted in a sharp rise in the pilot reactor temperature to just over 31^0C.

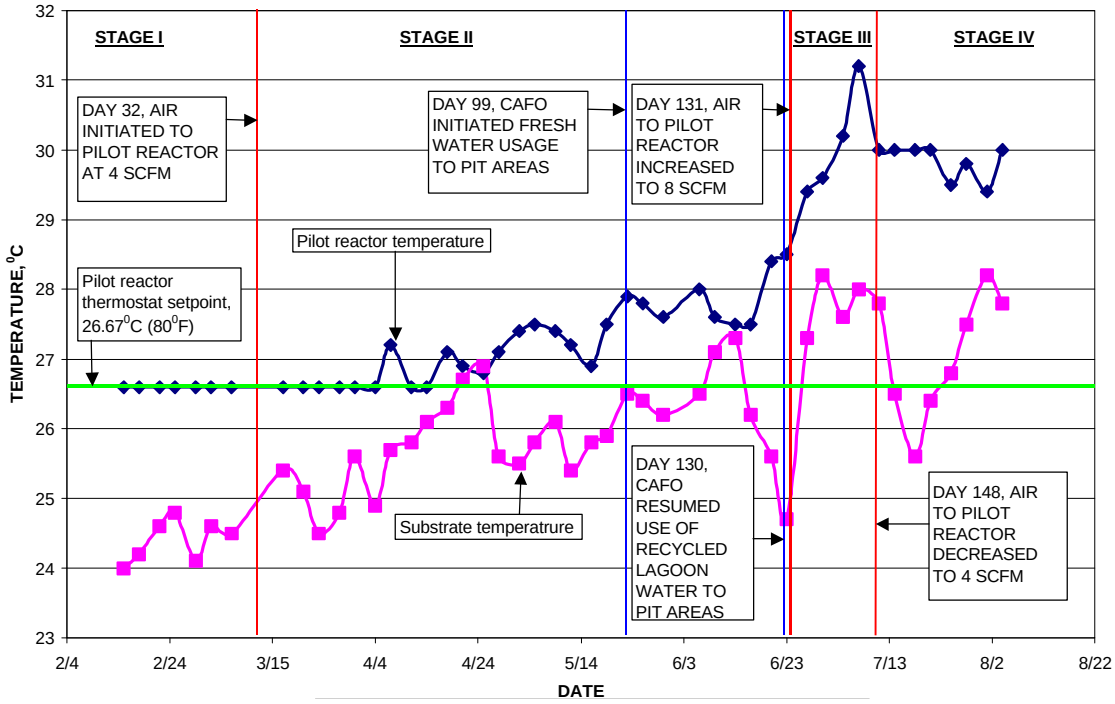

Figure 8.5 Pilot reactor temperature, substrate temperature, and stage profiles

Dissolved oxygen profile

In evaluating the success of the pilot reactor, the stratification of dissolved oxygen was important. Oswald (1994) indicated that dissolved oxygen levels for lightly loaded Advanced Facultative Ponds drop dramatically beginning at approximately three feet in depth and approach zero at approximately five feet in depth. Figure 8.6 shows the relationship of dissolved oxygen versus depth of the pilot reactor. The profile shown was determined during Stage II of the research. As shown, the air spargers were located at 2'-9" below the liquid surface. Above this elevation, the dissolved oxygen level was steady at 2.5 mg/L. Below two feet, the dissolved oxygen level dropped off dramatically to 0.35 mg/L and further decreased to 0.2 mg/L for the next three feet. The dissolved oxygen profile demonstrates proof of stratification for this parameter at this air-input rate.

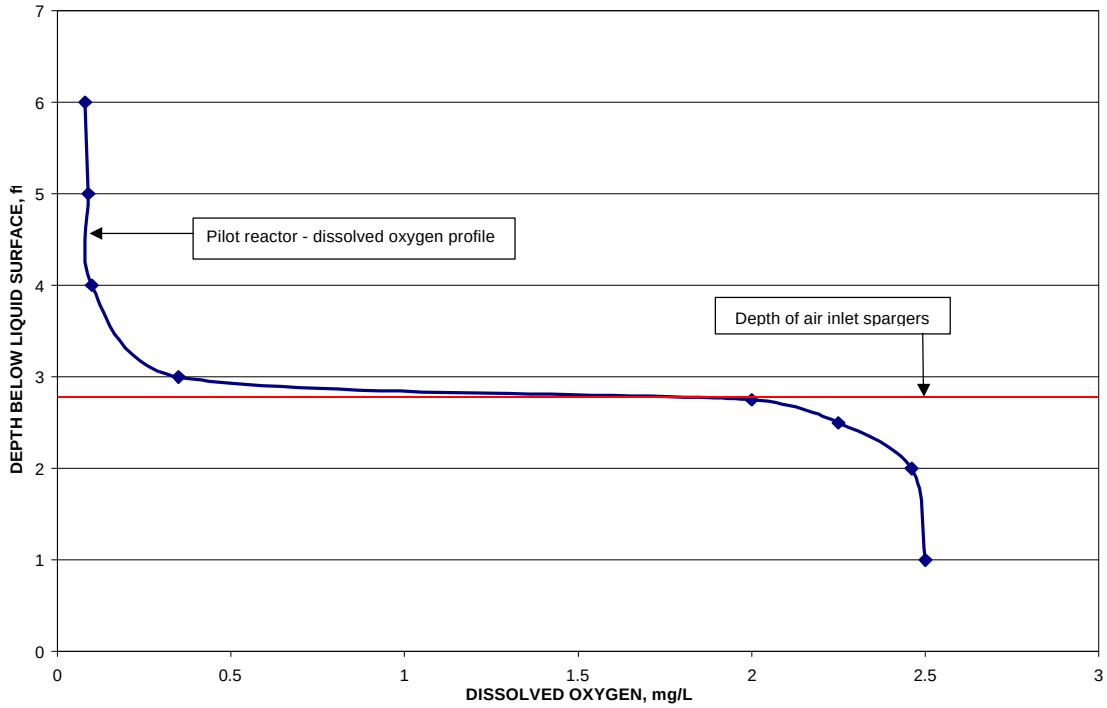

Figure 8.6 Pilot reactor – dissolved oxygen profile

Microbial populations

Chikh et al. (1997) demonstrated that, for a manure pond, the aerobic population ranges between $1.5x10^6$ CFU/mL to $2.65x10^7$ CFU/mL, and the anaerobic population ranges between $2.3x10^4$ CFU/mL to $9.4x10^4$ CFU/mL. In comparison, the pilot reactor yielded microbial populations capable of using oxygen as their terminal electron acceptor of $1x10^8$ CFU/mL in both the upper and lower strata. In addition, bacteria capable of using a terminal electron acceptor other than oxygen numbered $1x10^8$ CFU/mL in the lower strata, but dropped to $1x10^5$ CFU/mL in the upper strata. These data are shown graphically in Figure 8.7 and Figure 8.8. The data suggest that there may be a substantial population of facultative microorganisms involved. The differences in the populations found by Chikh versus those found from the pilot reactor in this project are attributed primarily to the fact that the pilot reactor was better controlled with regard to key parameters such as temperature, substrate loading, and dissolved oxygen levels.

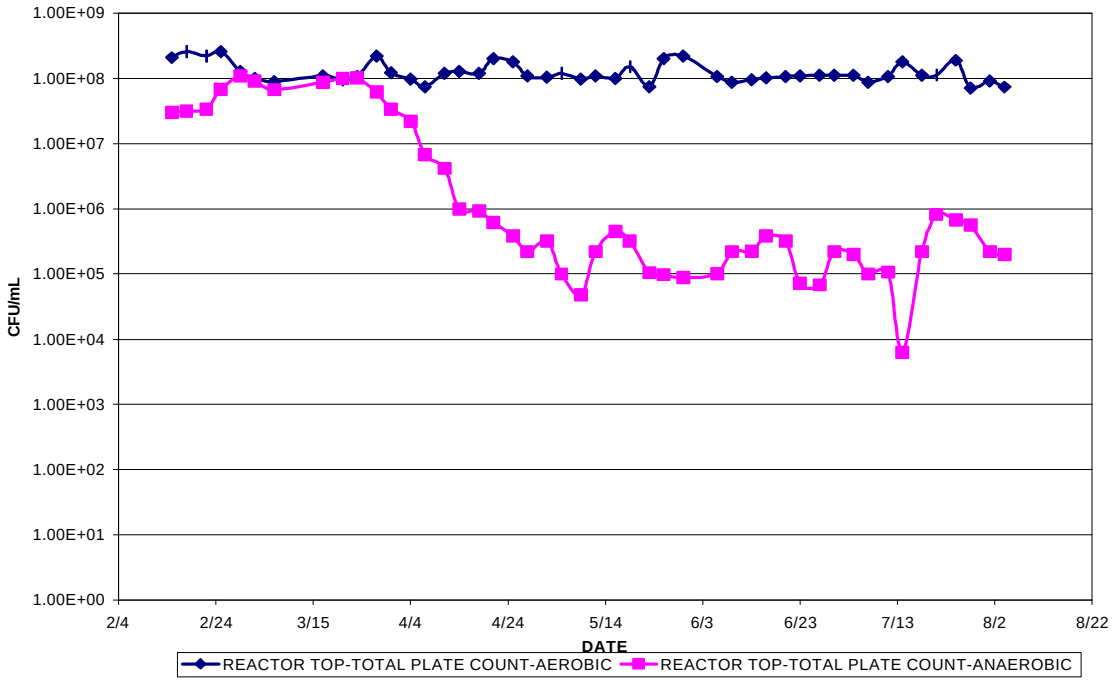

Figure 8.7 Pilot reactor – total microbial population – aerobic strata

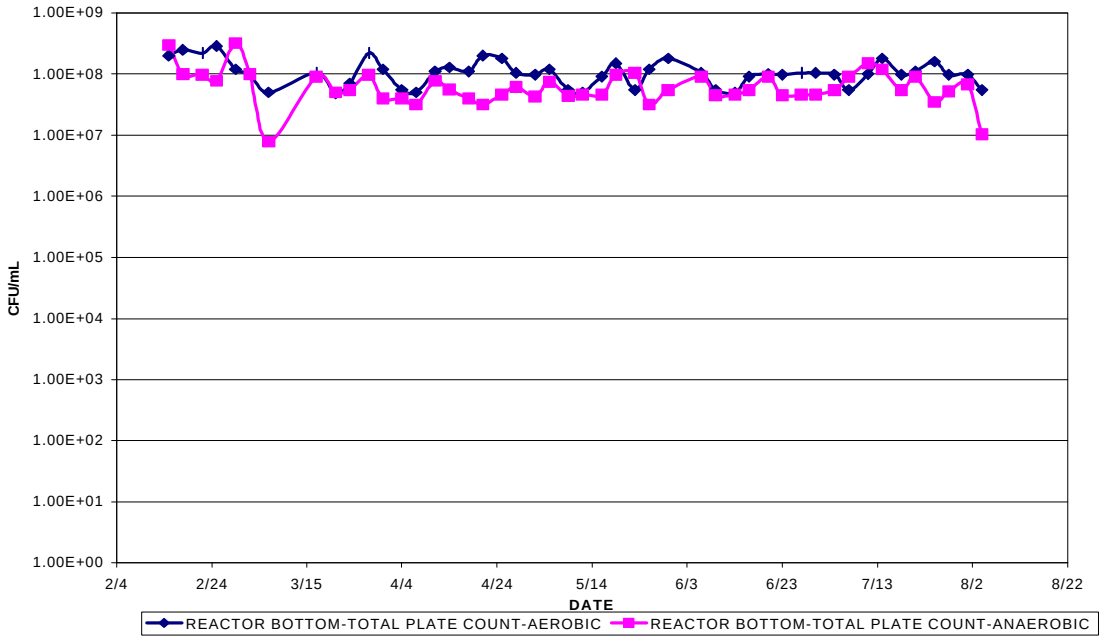

Figure 8.8 Pilot reactor – total microbial population – anaerobic strata

Odor reduction

Maintaining dissolved oxygen stratification to the extent that hydrogen-sulfide-producing bacteria and sulfate-reducing bacteria are unable to flourish in the upper strata was an objective of the project, thus reducing the opportunity for offensive odor production. In addition, the dissolved oxygen level had to be maintained high enough to overcome additional metabolic pathway requirements for degradation of organic materials reaching the upper strata, plus the oxidation of hydrogen-sulfide and mercaptan gases as they rise to surface. Figure 8.9 shows hydrogen-sulfide-producer populations and sulfate-reducer populations for the upper strata. As indicated, hydrogen-sulfide-producing bacteria existed at a level of $1x10^5$ CFU/mL during Stage I, but dropped dramatically approximately 20 day after the initiation of air into the reactor. And, with

the exception of one sample test result, the level of hydrogen-sulfide-producing bacteria remained below 10 CFU/mL for the life of the project. The sulfate-reducing bacteria, on the other hand, were never present in the upper strata where sufficient air was maintained at 4 SCFM to prevent the proliferation of hydrogen-sulfide-producing bacteria and sulfate-reducing bacteria. Recalling that hydrogen-sulfide gas has an odor index of 17,000,000 and a 100% odor recognition threshold concentration of 1 ppm (Verschueren, 1983), the fact that these bacteria were not present in the upper strata significantly reduces the possibility of odorous compounds of sulfur from reaching the atmosphere.

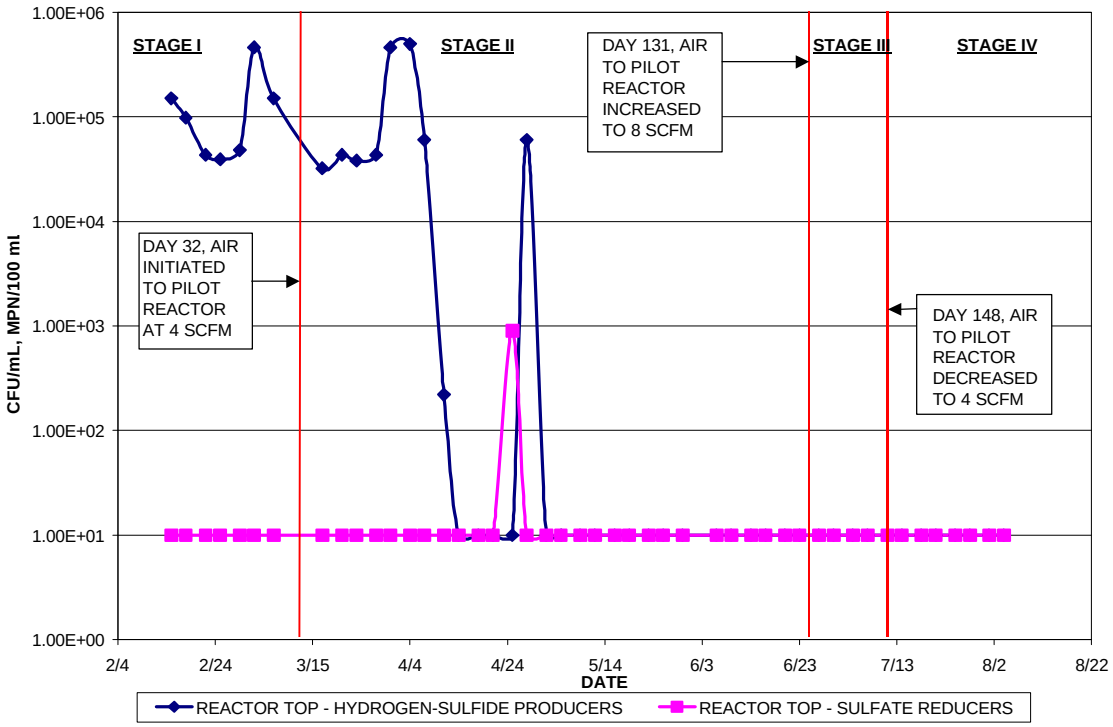

Figure 8.9 Hydrogen-sulfide producers and sulfate reducing microorganisms – aerobic strata

Motility and metabolic pathways

In addition to the overall microbial populations in the upper and lower strata, an examination of the microflora make-up was performed. This examination was necessary

to insure that each expected microbial type or metabolic process were present in sufficient numbers to perform the anticipated organic degradation. Figure 8.10 indicates the populations of hydrogen-sulfide producing organisms in the upper and lower strata. The average population in the aerobic strata was less than 10 CFU/mL, while the average population in the anaerobic strata averaged $1x10^5$ CFU/mL. These were to be expected since hydrogen-sulfide-producing bacteria thrive in anaerobic environments.

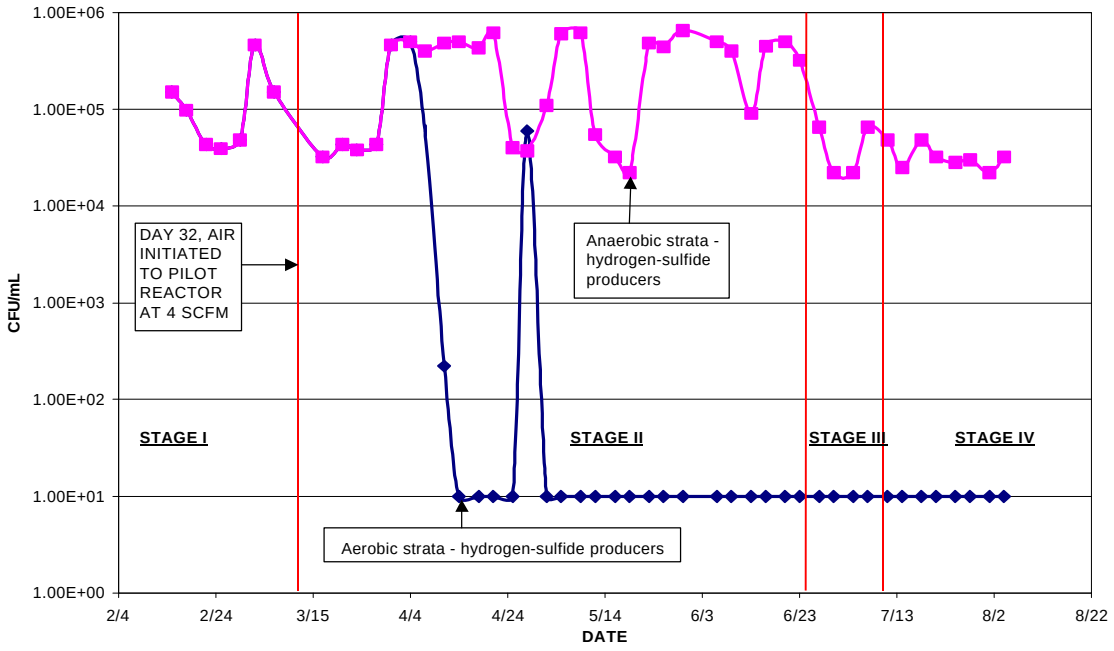

Figure 8.10 Microbial motility demonstrated by comparison of hydrogen-sulfide producing organisms in the aerobic and anaerobic strata

The graphic of the estimated number of sulfate-reducing bacteria in the pilot reactor is given in Figure 8.11. As shown, there were few sulfate-reducing bacteria present in the system at the onset of the project, but the bacteria grew in the anaerobic zone as the project moved forward, reaching a peak population of $1.2x10^4$ (MPN). There were never a significant number of sulfate-reducing bacteria in the upper strata. These results were also expected since sulfate-reducing organisms only thrive under anaerobic conditions.

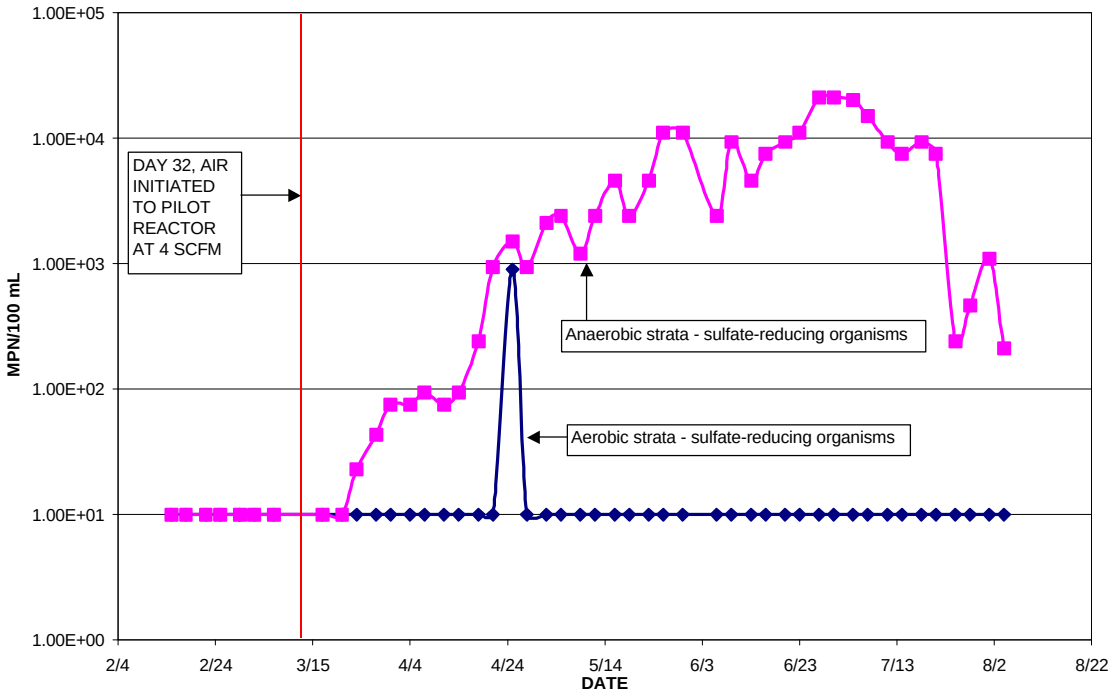

Figure 8.11 Microbial motility demonstrated by comparison of sulfate-reducing organisms in the aerobic and anaerobic strata

Bacterial populations responsible for reducing nitrate to nitrogen gas or ammonia can be seen in Figure 8.12. These denitrifiers flourished in both the aerobic and anaerobic strata at average estimated levels of $6.73x10^6$ (MPN) and $9.11x10^6$ (MPN), respectively.

The presence of denitrifiers in the lower strata is expected and explained by the fact that denitrification is a three-step process. The first two steps occur when the dissolved oxygen levels are above 1 mg/L, and the third step occurring under anoxic conditions. The numerous denitrifiers in the aerobic stratum were not expected. However, based on the fact that several type of heterotrophs are capable of reducing nitrate to nitrogen gas may explain why the number of denitrifying organisms are this high in magnitude.

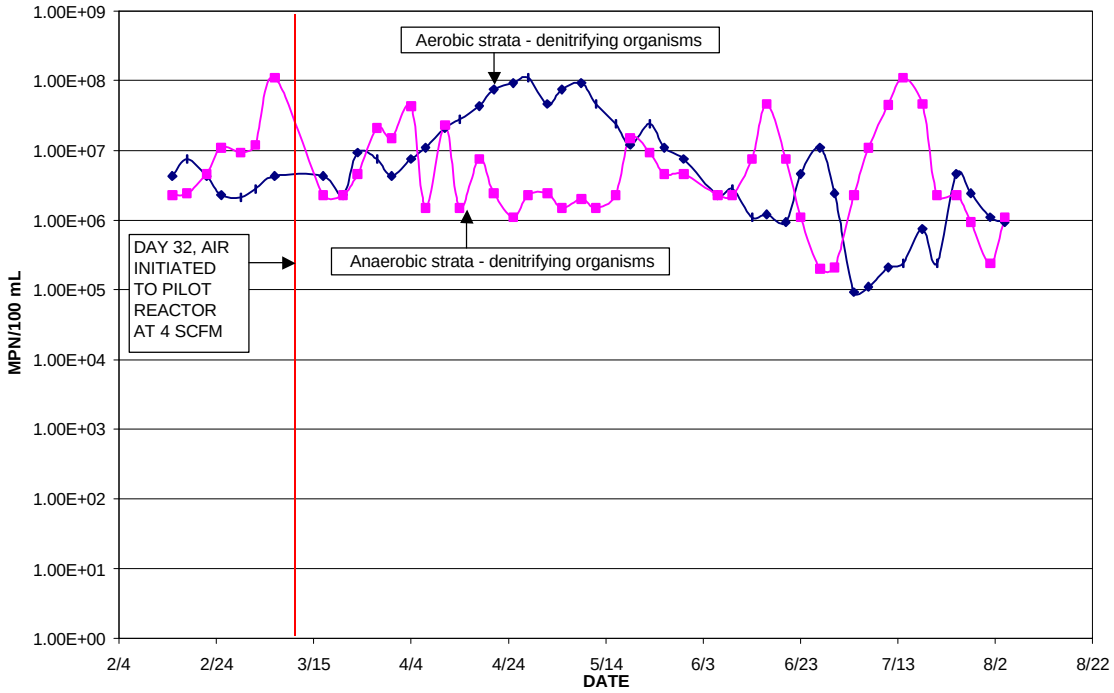

Figure 8.12 Microbial motility demonstrated by comparison of denitrifying organisms in the aerobic and anaerobic strata

The estimated number of microorganisms capable of producing acid and gas from glucose as their carbon source are shown in Figure 8.13. This graphic shows the estimated number of carbohydrate-utilizers for both the aerobic and anaerobic strata. These estimated populations were 6.2×10^6 (MPN) and 7.2×10^6 (MPN) in the aerobic and anaerobic strata, respectively. Because carbohydrates are a part of the animals' diet, it

was expected that there would be a significant number of carbohydrate-utilizers in both strata.

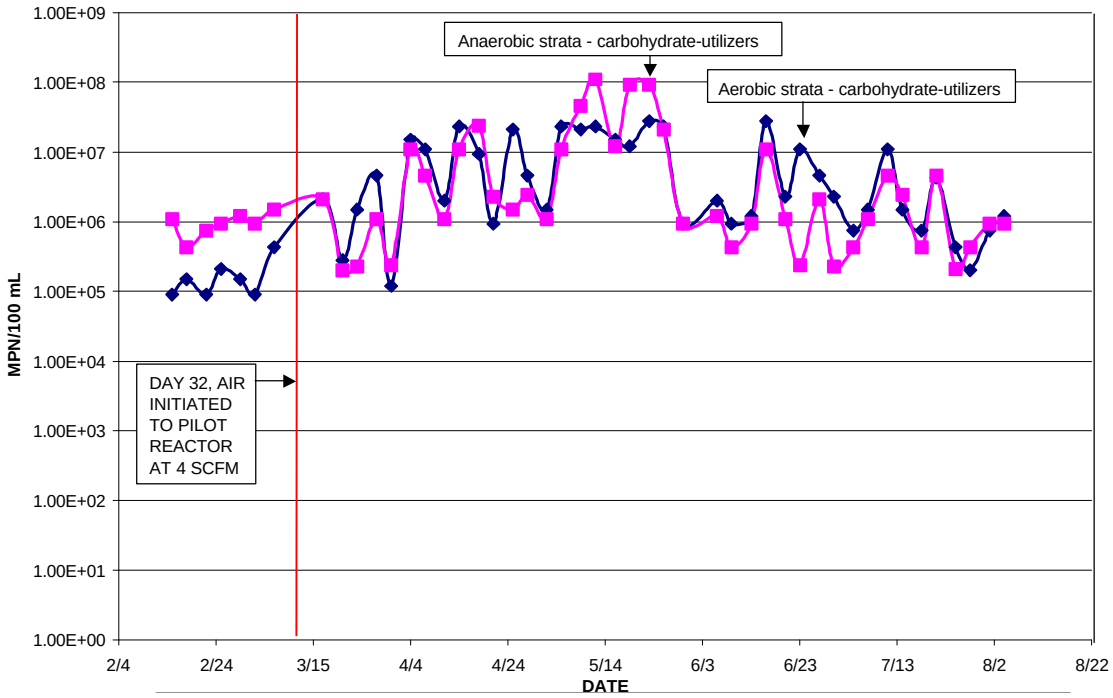

Figure 8.13 Microbial motility demonstrated by comparison of carbohydrate-utilizers in the aerobic and anaerobic strata

In summary, the aerobic stratum was found to have significant numbers of denitrifying bacteria and carbohydrate-utilizing bacteria. The anaerobic strata was also found to have significant numbers of denitrifying and carbohydrate-utilizing bacteria, plus a significant number of bacteria capable of reducing sulfate and bacteria capable of producing hydrogen-sulfide. These data support the fact that microoganisms move toward environments more favorable to their needs and requirements, and away from those environments that are otherwise toxic to them. In addition, the data support the fact that the degradation activities occurring in the pilot reactor were not a result of differences in the microbial populations, but rather is attributed to difference in the

metabolic pathways being employed, although some bacteria are only active in one mode, either aerobic or anaerobic.

pH and carbohydrate-acid producers

From the initiation of air into the pilot reactor on day 32 of the project, there was a steady rise in the pH of the system. The pH in the upper and lower strata tracked closely together, with a Pearson coefficient of correlation of 0.98. In addition, the data indicated a fairly consistent population level of carbohydrate-utilizing bacteria that produce acid as a part of their metabolic process. This fact was true of both the aerobic and anaerobic strata. These estimated populations, as previously noted, were $6.2x10^6$ (MPN) and $7.2x10^6$ (MPN) on average for the upper and lower strata, respectively. Figure 8.14 shows the graphic for pH and acid-producing carbohydrate-utilizers in the aerobic strata and Figure 8.15 shows the same graphic for the anaerobic strata. As indicated, the pH rose from approximately 7.7 to 9.0 during the project life. These data seem to conflict with one another. However, this seeming contradiction can be explained by the fact that there must exist a significant number of protein utilizers in the system as well. And, since the hog's diet is one of protein-enriched feedstock, it is reasonable to assume that the protein utilizers are controlling the pH and not the carbohydrate utilizers that produce acid.ı

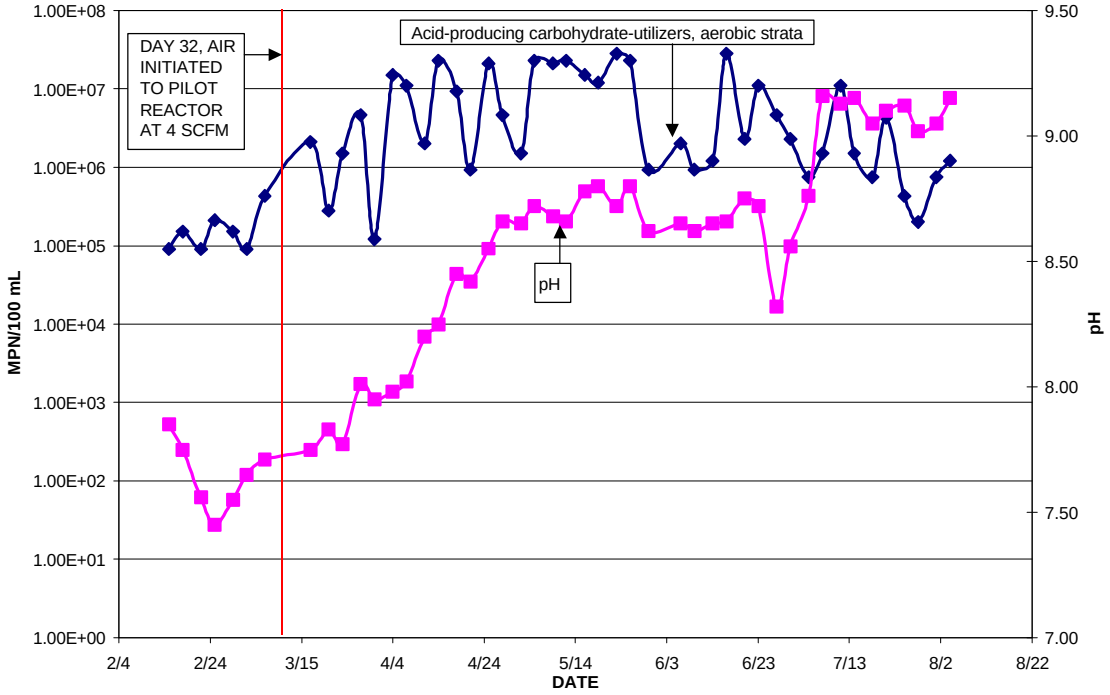

Figure 8.14 Acid-producing carbohydrate-utilizers in the aerobic strata

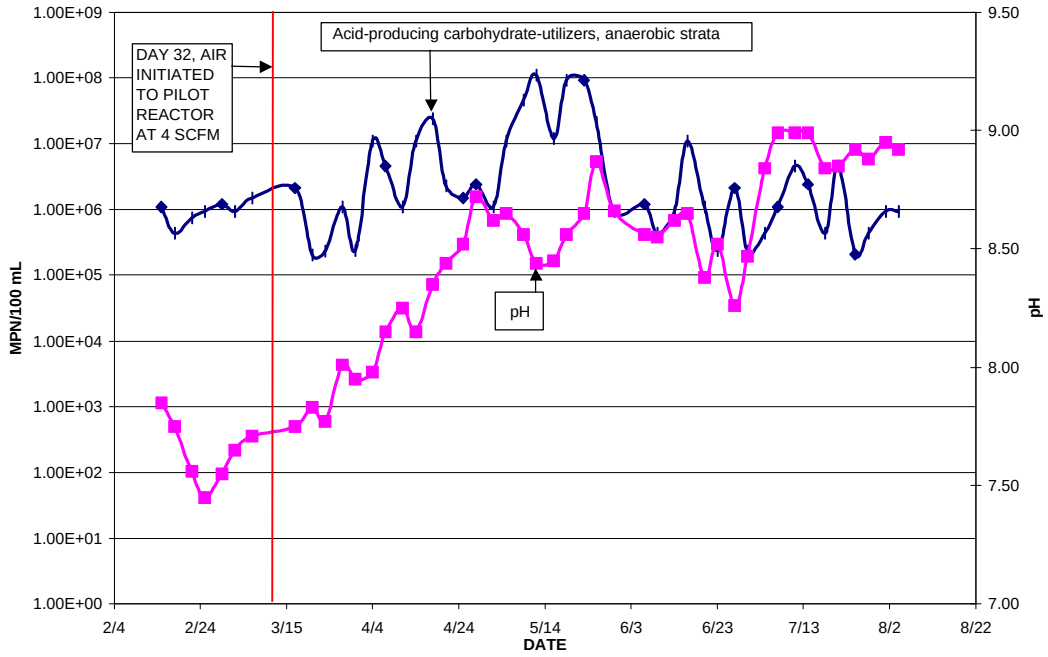

Figure 8.15 Acid-producing carbohydrate-utilizers in the anaerobic strata

Biochemical oxygen demand

Throughout the project the biochemical oxygen demand was closely monitored. Figure 8.16 shows the BOD_5 for the substrate, the pilot reactor lower strata, and pilot reactor upper strata. On average these values were 367 mg/L, 94.2 mg/L, and 77.2 mg/L, respectively. The graphic clearly demonstrates a consistent BOD_5 reduction of 75% overall, with 18% as a result of aerobic treatment. The fact that 18% is attributed to the upper strata is significant since the system must insure that an adequate amount of air is introduced to system for the additional degradation occurring in the upper strata.

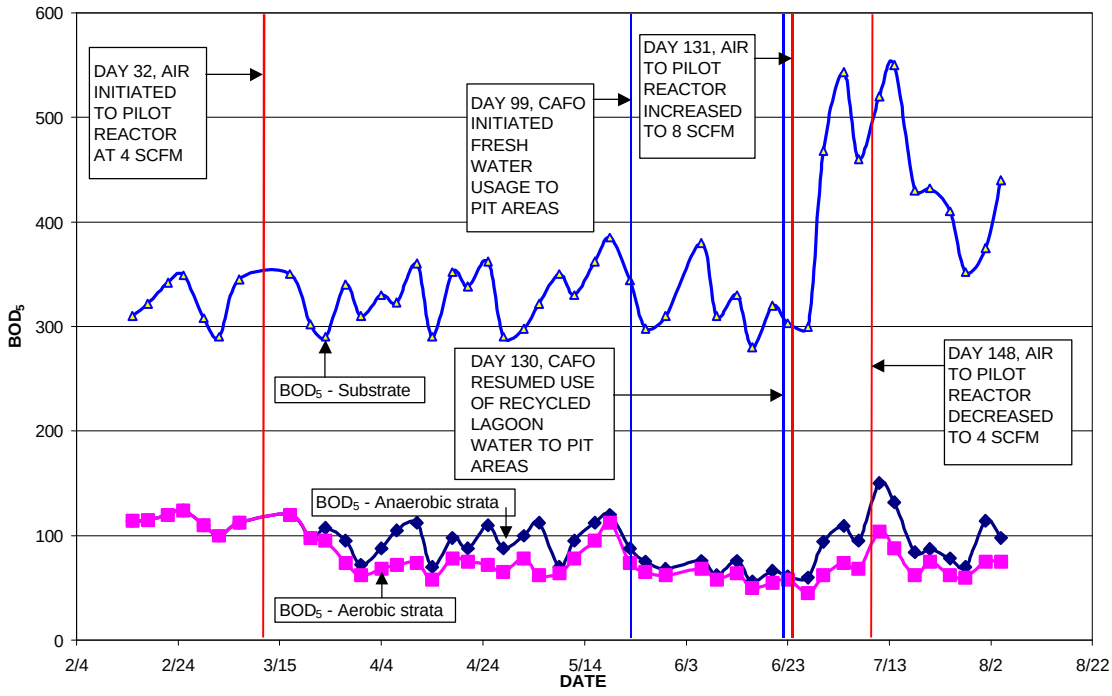

Figure 8.16 Biochemical oxygen demands for substrate, anaerobic strata, and aerobic . . . strata

Solids

The total suspended solids and volatile suspended solids data is shown graphically in Figure 8.17. The average total suspended solids in the upper strata were 687 mg/L and 598 mg/L in the lower strata. The average volatile suspended solids were 334 mg/L in the upper strata and 328 mg/L in the lower strata. Based on these data, the volatile suspended solids represented 49% of the total suspended solids in the upper strata, and 55% of the total suspended solids in the lower strata.

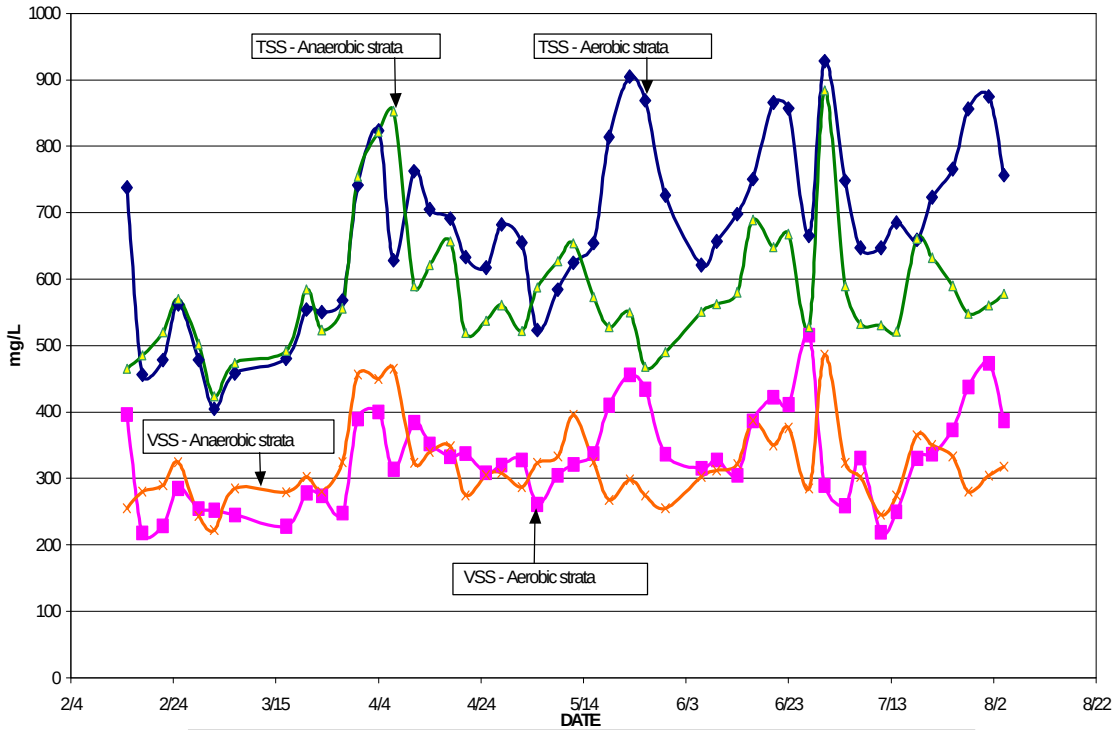

Figure 8.17 Total suspended solids and volatile suspended solids for aerobic and anaerobic strata

In addition, an analysis was performed on solids accumulation and sludge formation. Upon completion of the project, the reactor was slowly drained and the solids in the bottom of the reactor were collected. These solids were tested to determine the fixed and volatile parts. These results are shown in Table 8.1.

Table 8.1 Pilot Reactor – Solids Production

Parameter	Result
1. Total Solids (wet)	40.1 L (approximately 10 gallons)
2. Total Fixed Solids	2,590 g (5.7 lbs)
3. Total Volatile Solids	33,106 g (73.9 lbs)

Through the course of the experiment, 350 gallons of substrate samples were collected and introduced into the reactor. Assuming no fixed or volatile solids in the effluent, this would result in 1,955 mg of fixed solids/L of wastewater and 25,990 mg of volatile solids/L of wastewater.

Additional Observations

In addition to those specific observations previously mentioned, all data gathered throughout the project were tabled and plotted as a single parameter versus time. These data and plots can be seen for the wastewater substrate, the anaerobic strata, and the aerobic strata in Appendices A, B, and C, respectively.

In addition, the data were correlated with regard to each parameter for the upper and lower strata. These data were then plotted against time. Table D-1 in Appendix D shows this data, and Table D-2 shows the Pearson correlation coefficients for each data set.

Plots of the data are found in Figures D.1 through D.16. As seen from the Pearson correlation coefficients the following data sets were strongly correlated: total aerobic plate counts, ammonia-nitrogen, nitrate, sulfate, sulfide, pH, temperature, and BOD. Those data sets that showed little correlation included denitrifiers, sulfate reducers, and volatile suspended solids.

Appendix E shows correlated data from the influent wastewater to the pilot reactor, with the effluent wastewater from the upper strata of the reactor. The data are found in Table E-1 and the Pearson correlation coefficients are shown in Table E-2. Plots of the data are found in Figures E.1 through E.17. Those data sets that demonstrated a strong correlation included ammonia-nitrogen and temperature. Those data sets that showed little correlation included total aerobic plate counts, total anaerobic plate counts, hydrogen-sulfide producers, carbohydrate-utilizers (gas and acid producers), denitrifiers, sulfate reducers, dissolved oxygen, BOD_5, total suspended solids, and volatile suspended solids.

CHAPTER IX

CONCLUSIONS AND RECOMMENDATIONS

With the increasing concern regarding how and where livestock are grown, more debate and regulation can be expected. As the current research continues on environmental and odor issues surrounding CAFOs, opponents of these facilities also continue to apply pressure to owners and regulators. A great amount of research has already been performed on a wide variety of issues relating to CAFOs, but as of now, there is no consensus on a facility design that pleases all stakeholders. It was the goal of this research to attempt to address a part of the concerns related to the swine CAFO.

The intent of this research was to simulate as closely as possible a full-scale operation. Thus, it was essential to fabricate a reactor of adequate depth to provide the environment necessary for stratification of the microflora if it were, in fact, to occur. However, for several reasons, the reactor depth was limited to only 8 feet. For a full-scale design, the lagoon would more likely be 15 to 20 feet deep.

In addition, although all materials came from an actual CAFO, the method of air injection into the reactor was strictly a bench scale method. In a full scale design the method of air introduction would have been modified to adequately satisfy the objectives.

Conclusions

Based on the results of this investigation, several conclusions have been drawn about the effectiveness of the biological degradation, the microbial mechanisms of the process, and the stratification of the treatment system. These observations yield insight into the potential for future implementation of this process for CAFO designs. The major points of this analysis are as follows:

1. The liquid in the pilot reactor treatment system was stratified in regard to oxygen content with the upper 2.75 feet being aerobic and the bottom 4.75 feet being anaerobic.
2. The overall microbial population remained consistent at $1x10^8$ CFU/mL in the upper and lower strata of the pilot reactor.
3. Hydrogen-sulfide-producing organisms and sulfate-reducing organisms were non-detectable in the upper zone of the reactor in Stage II of the process, thus reducing the potential to release offensive odorous compounds into the atmosphere.
4. Differences in substrate decomposition were due to differences in the metabolic pathways employed by the microflora (aerobic vs. anaerobic) rather than changes in the microbial populations.
5. The data support the observation that microorganisms move toward more favorable environments and away from those areas that may be toxic to them. This fact is evident in the number of hydrogen sulfide producers, sulfate reducers, and denitrifiers found in the anaerobic zone of the reactor.
6. The steady rise in pH throughout the experiment appeared to conflict with the number of acid-producing carbohydrate-utilizers found. However, this seeming

conflict can be explained by the fact that protein metabolism (ammonification) contributes to a rise in pH, thus offsetting acid production. Also, the utilization of the organic acids by other microorganisms would decrease the H^+ ion concentration.

7. The 5-day, 20^0C biochemical oxygen demand reduction overall was 75%, with 18% of that reduction attributed to the aerobic zone.
8. Data obtained at the end of the process cycle revealed 1,955 mg of fixed solids per liter of wastewater and 25,990 mg of volatile solids per liter of wastewater.

In summary, the adaptation of an upflow anaerobic/aerobic treatment system for handling swine wastewaters to accomplish specific odor related reductions is possible. Noting that the addition of the aerobic zone is primarily for the oxidation of odorous compounds, and not for the enhanced biological degradation, the amount of oxygen required can be minimized. Furthermore, changes in the operational procedures of the CAFO with regard to draining of the holding structures also may have an impact on the oxygen requirement. A more consistent loading of the lagoon system would yield a more uniform substrate and move the system toward a continuous flow system instead of quasi-batch system.

Recommendations

This project raises several issues, which should undergo further examination.

1. There are indications that protein-utilizing microorganisms are causing an increase in pH due to ammonification. An examination into these indications, along with an examination of the hog's diet would shed additional light on this issue.

2. This study did not examine the oxidation/reduction potentials that existed throughout the pilot reactor. An evaluation of these potentials would be valuable in understanding both the development, activity, and metabolic pathways of the microbial population.
3. Additional work is needed to quantify the odor reduction obtained utilizing the waste treatment process employed in this project. This work should include head-space analysis and odor sensory evaluations.
4. The best method of introducing oxygen into a CAFO lagoon requires additional research. There are many methods which have been researched and tested on similar systems, but there still exists opportunity to confirm the best method for accomplishing the unique objective of odor reduction and stratification of anaerobic and aerobic zones, since this study suggests that changes in metabolic pathways rather than changes in the composition of the microbial populations are involved.
5. Full-scale testing would confirm the validity of the assumptions made in this study such as the air sparging depth. Full-scale testing would also provide a better understanding of the dissolved oxygen profile in all directions and help to determine the size and number of air spargers.
6. A longer study would also help to confirm the results of this study. Looking at each parameter for an entire 12-month cycle would assist in understanding the microbial activity and any problems that may arise therein.
7. A controlled study to fully understand the effects of temperature variation on the microbial population would be beneficial to the full understanding of this system.

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APPENDIX A

SUBSTRATE DATA ANALYSIS

Table A.1 Substrate (CAFO) House Discharge) – Data Analysis

PARAMETER	3/16	3/19	3/23	3/26	3/30	3/9	3/7	3/17	3/31	3/24	3/28	3/31	4/4	4/7	4/11	4/14	4/18	4/21	4/25	4/2	4/9	4/
SUBSTRATE-TOTAL PLATE COUNT-AEROBIC	1.23E+07	6.80E+07	1.10E+07	1.50E+07	4.80E+07	9.60E+07	1.00E+07	1.10E+07	1.10E+07	1.50E+07	1.10E+08	2.30E+08	1.50E+07	1.80E+07	5.30E+07	1.10E+07	4.90E+07	9.30E+07	1.23E+07	2.30E+07	1.00E+07	1.00E+07	2.30E+08
SUBSTRATE-TOTAL PLATE COUNT-ANAEROBIC	2.30E+06	3.80E+07	3.10E+08	8.00E+08	9.20E+08	3.75E+07	6.00E+06	6.00E+06	5.80E+08	1.10E+07	3.20E+07	1.10E+08	4.80E+08	9.80E+08	3.90E+07	6.80E+08	3.90E+07	7.50E+07	1.50E+07	1.80E+07	5.80E+08	7.20E+08	1.90E+07
SUBSTRATE-HYDROGEN SULFIDE PRODUCERS	2.10E+04	3.20E+04	3.90E+04	8.00E+04	5.20E+04	3.00E+04	5.50E+04	4.80E+05	9.00E+05	4.40E+05	4.80E+05	3.20E+05	9.00E+05	6.80E+05	4.80E+05	3.20E+05	6.90E+05	1.00E+05	7.80E+05	3.20E+05	9.50E+04	8.50E+04	4.80E+04
SUBSTRATE-CARBOHYDRATE UTILIZERS ACID PRODUCERS	1.10E+07	4.80E+06	1.10E+07	2.10E+06	4.30E+06	9.30E+05	4.30E+06	2.00E+06	7.80E+06	1.10E+07	1.10E+07	4.60E+06	1.10E+07	2.10E+06	2.30E+06	2.10E+06	1.10E+07	1.20E+06	1.10E+07	1.10E+07	1.10E+07	4.60E+06	1.10E+07
SUBSTRATE-CARBOHYDRATE UTILIZERS GAS PRODUCERS	1.10E+07	4.80E+06	1.10E+07	2.10E+06	4.30E+06	9.30E+05	4.30E+06	2.00E+06	7.80E+06	1.10E+07	4.60E+06	4.60E+06	1.10E+07	2.10E+06	2.30E+06	2.10E+06	1.10E+07	1.20E+06	1.10E+07	1.10E+07	1.10E+07	4.60E+06	2.10E+06
SUBSTRATE-DENITRIFERS	2.60E+06	6.40E+06	7.90E+05	1.20E+06	1.10E+07	4.60E+05	1.10E+06	4.60E+06	2.40E+07	2.90E+06	2.30E+06	1.20E+07	1.50E+07	1.80E+06	2.10E+06	1.10E+06	4.90E+07	2.10E+06	4.60E+06	2.10E+07	4.30E+06	4.30E+06	9.30E+05
SUBSTRATE-SULFATE REDUCERS	7.60E+03	1.50E+04	2.10E+04	1.10E+06	2.10E+06	4.60E+05	1.10E+05	2.10E+06	9.30E+06	7.80E+06	1.10E+06	4.60E+05	2.60E+04	4.30E+04	1.50E+04	7.50E+04	9.30E+03	7.50E+03	2.00E+04	7.50E+04	9.30E+04	2.10E+04	2.60E+04
SUBSTRATE-AMMONIA-N	1025		980			400			366		270				690				880				400
SUBSTRATE-NITRATE	0.0		56.0			42.0			86.0		42.0				17.6				160.0				42.0
SUBSTRATE-SULFATE	104.0		206.0			78.0			116.0		56.5				178.0				58.0				96.0
SUBSTRATE-SULFIDE	0.9		0.0			1.6			0.0		0.0				0.0				0.0				0.8
SUBSTRATE-pH	7.84	7.76	7.21	6.98	6.87	7.14	7.82	7.12	7.24	7.86	7.11	6.99	6.85	7.01	7.14	7.24	7.18	7.84	7.80	7.32	7.14	7.36	7.00
SUBSTRATE-DISSOLVED OXYGEN	0.05	0.06	0.04	0.05	0.04	0.05	0.03	0.04	0.02	0.02	0.06	0.06	0.07	0.06	0.03	0.04	0.02	0.02	0.03	0.06	0.04	0.06	0.06
SUBSTRATE-TEMPERATURE	24.0	24.2	24.8	24.8	24.1	24.8	24.5	25.6	25.1	24.8	24.8	25.8	24.9	25.7	25.6	25.1	26.3	25.7	25.9	25.6	25.5	25.8	26.1
SUBSTRATE-BOD	310	322	342	349	306	290	345	350	302	290	340	310	320	323	360	290	352	334	362	290	298	322	350
SUBSTRATE-TSS	550	640	632	640	1120	930	1020	810	440	670	636	811	1170	960	740	705	860	815	730	710	730	1040	860
SUBSTRATE-VSS	445	560	496	520	923	705	520	680	366	536	496	758	940	730	486	560	708	968	452	620	686	852	813

PARAMETER	8/12	8/16	8/19	8/23	8/26	8/30	8/9	8/9	8/13	8/16	8/23	8/23	8/27	8/30	7/6	7/7	7/11	7/14	7/18	7/21	7/28	7/28	8/1	8/4
SUBSTRATE-TOTAL PLATE COUNT-AEROBIC	9.50E+07	4.60E+07	6.90E+07	9.90E+07	7.80E+07	1.10E+07	3.20E+07	1.20E+07	1.00E+07	3.30E+07	1.00E+07	4.00E+07	3.20E+07	5.50E+07	7.80E+07	1.10E+07	3.80E+07	3.80E+07	9.90E+07	7.00E+07	5.20E+07	3.20E+07	1.20E+07	1.10E+07
SUBSTRATE-TOTAL PLATE COUNT-ANAEROBIC	5.50E+07	2.20E+07	7.80E+07	6.20E+07	4.80E+07	9.20E+06	2.00E+06	6.40E+06	3.20E+06	6.00E+06	4.80E+06	6.00E+06	5.00E+06	9.00E+06	4.00E+07	6.00E+06	8.00E+06	1.10E+07	4.50E+07	3.60E+07	4.00E+07	1.50E+07	7.80E+06	6.80E+06
SUBSTRATE-HYDROGEN SULFIDE PRODUCERS	6.90E+04	5.60E+04	2.10E+04	3.00E+04	5.60E+04	3.90E+05	3.10E+04	9.00E+04	9.00E+04	1.00E+05	6.00E+04	9.20E+05	3.20E+05	4.40E+05	5.00E+05	9.00E+04	7.60E+05	3.00E+05	8.20E+04	9.20E+04	2.10E+04	5.00E+04	3.00E+04	3.80E+04
SUBSTRATE-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+07	1.10E+07	1.10E+07	4.60E+06	1.10E+07	1.60E+06	2.10E+06	1.50E+06	2.40E+06	2.40E+06	4.60E+06	7.80E+06	1.10E+07	1.10E+07	4.60E+06	1.10E+07	1.10E+07	2.40E+06	1.10E+07	1.10E+07	1.10E+07	4.60E+06	1.10E+07	1.10E+07
SUBSTRATE-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	1.10E+07	1.10E+07	1.10E+07	4.60E+06	1.10E+07	1.60E+06	2.10E+06	1.50E+06	2.40E+06	2.40E+06	4.60E+06	7.80E+06	1.10E+07	4.60E+06	4.60E+06	1.10E+07	1.10E+07	2.40E+06	1.10E+07	2.40E+06	1.10E+07	4.60E+06	1.10E+07	1.10E+07
SUBSTRATE-DENITRIFERS	1.10E+07	2.60E+06	2.60E+06	7.60E+06	9.30E+06	2.10E+06	4.60E+06	1.50E+06	4.30E+06	2.60E+06	2.10E+06	4.60E+06	9.30E+06	1.20E+07	1.60E+07	7.60E+06	9.30E+06	1.60E+07	2.10E+07	4.30E+06	2.60E+06	1.50E+06	9.30E+06	4.30E+06
SUBSTRATE-SULFATE REDUCERS	1.10E+06	1.60E+04	1.50E+04	1.10E+06	2.40E+04	9.30E+04	1.60E+04	1.50E+04	9.30E+04	4.30E+06	9.30E+06	7.60E+03	1.20E+04	4.60E+06	1.10E+06	2.40E+06	1.50E+06	2.40E+06	1.10E+06	1.10E+06	4.60E+06	2.40E+06	2.40E+06	9.30E+04
SUBSTRATE-AMMONIA-N									580				430			290				860				
SUBSTRATE-NITRATE									62.0				140.0			34.0				300.0				
SUBSTRATE-SULFATE									220.0				122.0			78.4				66.5				
SUBSTRATE-SULFIDE									0.0				0.0			0.0				1.4				
SUBSTRATE-pH	7.48	7.65	7.30	7.21	6.99	6.92	7.11	7.32	7.86	7.84	7.82	7.79	7.21	6.98	6.87	7.14	7.82	7.24	7.51	7.82	7.84	7.75	7.81	7.41
SUBSTRATE-DISSOLVED OXYGEN	0.05	0.05	0.04	0.05	0.76	0.51	0.52	0.68	0.66	0.75	0.81	0.84	0.48	0.04	0.02	0.06	0.05	0.06	0.04	0.04	0.05	0.06	0.04	0.03
SUBSTRATE-TEMPERATURE	26.4	26.8	26.9	26.6	26.4	26.2	26.8	27.1	27.3	26.2	25.6	24.7	27.3	28.2	27.8	26.0	27.5	26.6	25.6	26.4	26.8	27.5	26.2	27.6
SUBSTRATE-BOD	330	362	366	344	286	310	360	310	330	280	330	303	300	466	543	480	520	550	430	432	410	362	370	440
SUBSTRATE-TSS	960	900	560	940	1130	1290	660	802	560	427	526	470	760	910	1182	1310	947	870	660	626	960	1120	780	960
SUBSTRATE-VSS	784	668	445	525	806	782	720	490	433	345	486	382	626	766	805	1060	770	706	580	415	748	868	642	720

Table A.2 Substrate Characterization Plots

FIGURES A.1 THROUGH A.17		
A.1	SUBSTRATE	TOTAL PLATE COUNT - AEROBIC
A.2	SUBSTRATE	TOTAL PLATE COUNT - ANAEROBIC
A.3	SUBSTRATE	HYDROGEN-SULFIDE PRODUCERS
A.4	SUBSTRATE	CARBOHYDRATE-UTILIZERS - ACID PRODUCERS
A.5	SUBSTRATE	CARBOHYDRATE-UTILIZERS - GAS PRODUCERS
A.6	SUBSTRATE	DENITRIFIERS
A.7	SUBSTRATE	SULFATE REDUCERS
A.8	SUBSTRATE	AMMONIA-NITROGEN
A.9	SUBSTRATE	NITRATE-NITROGEN
A.10	SUBSTRATE	SULFATE
A.11	SUBSTRATE	SULFITE
A.12	SUBSTRATE	pH
A.13	SUBSTRATE	DISSOLVED OXYGEN
A.14	SUBSTRATE	BOD
A.15	SUBSTRATE	TOTAL SUSPENDED SOLIDS
A.16	SUBSTRATE	VOLATILE SUSPENDED SOLIDS
A.17	SUBSTRATE	TEMPERATURE

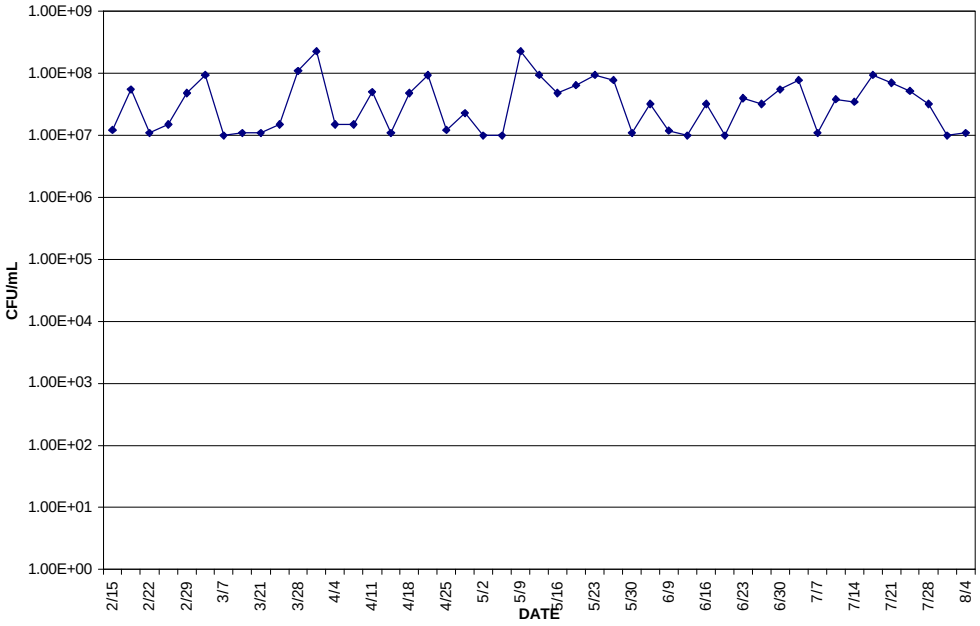

Figure A.1 Substrate – total plate counts - aerobic

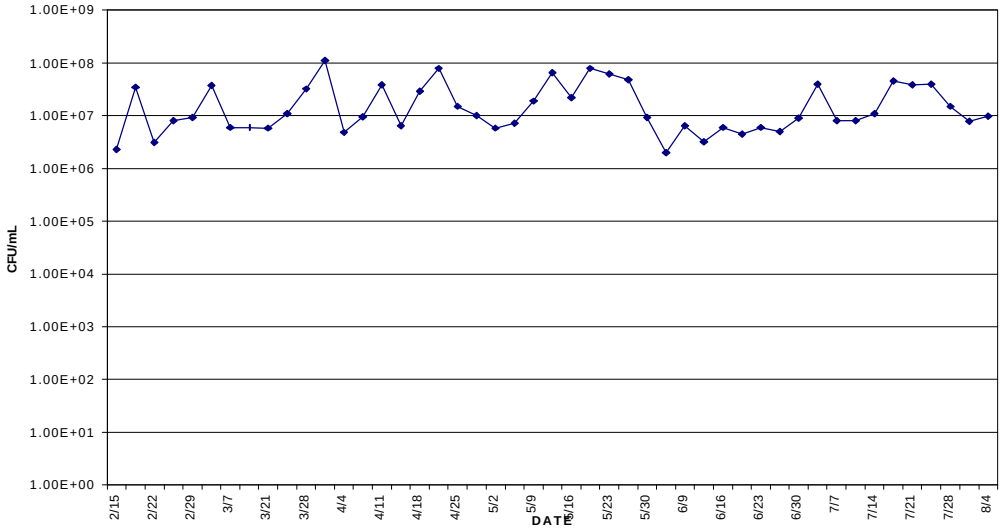

Figure A.2 Substrate – total plate count - anaerobic

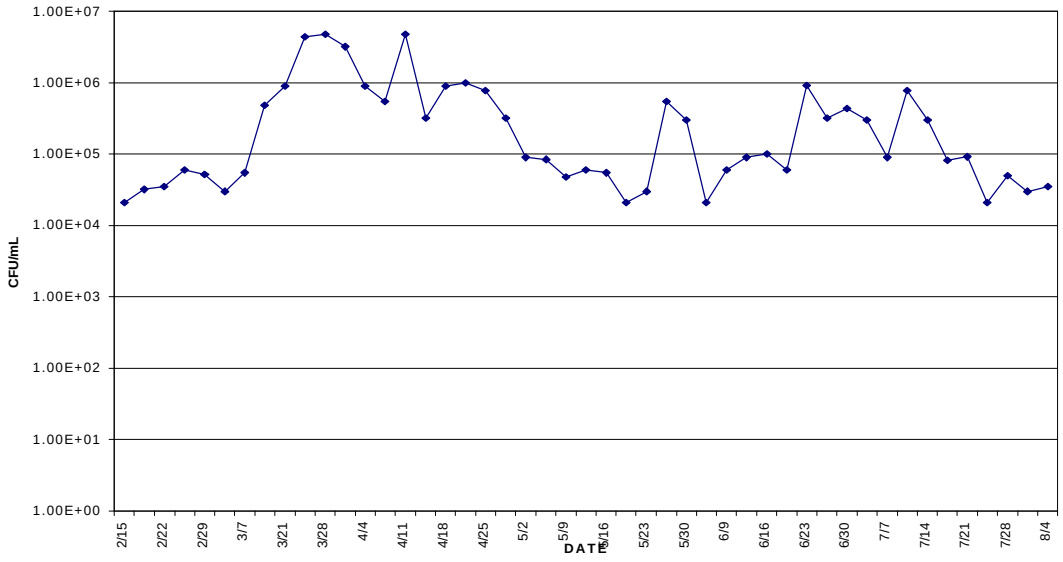

Figure A.3 Substrate – hydrogen-sulfide producers

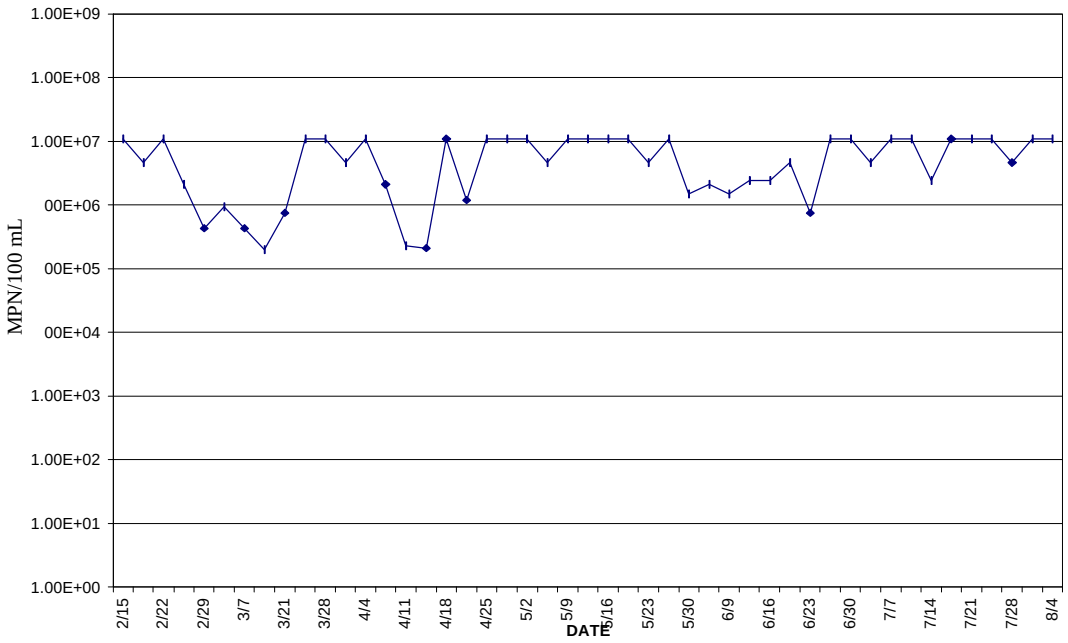

Figure A.4 Substrate – carbohydrate-utilizers – acid producers

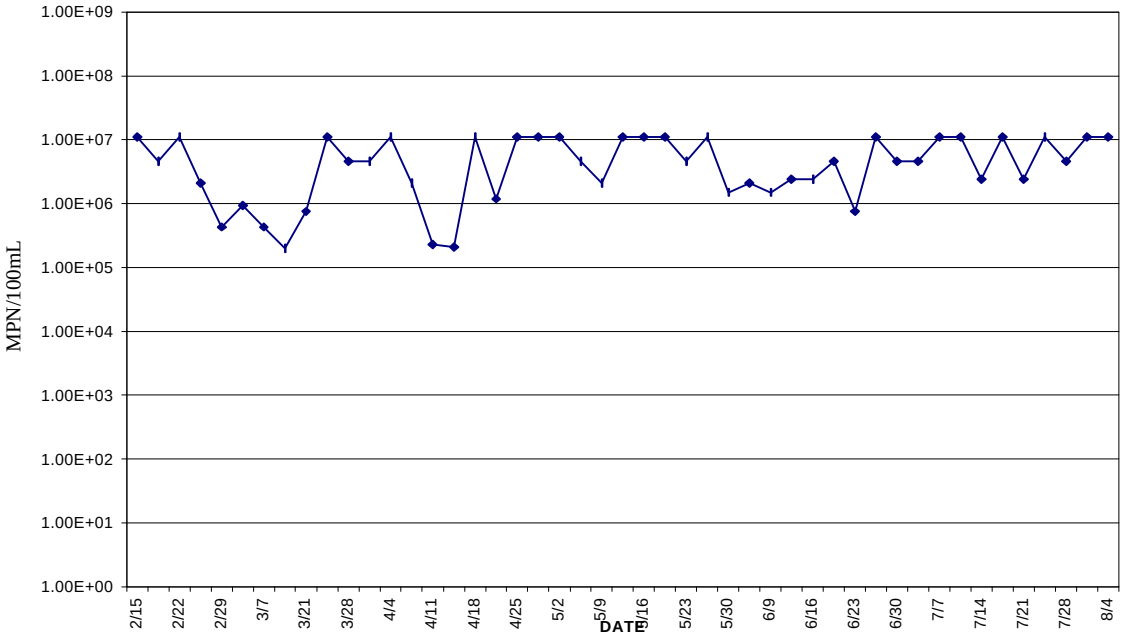

Figure A.5 Substrate – carbohydrate-utilizers – gas-producers

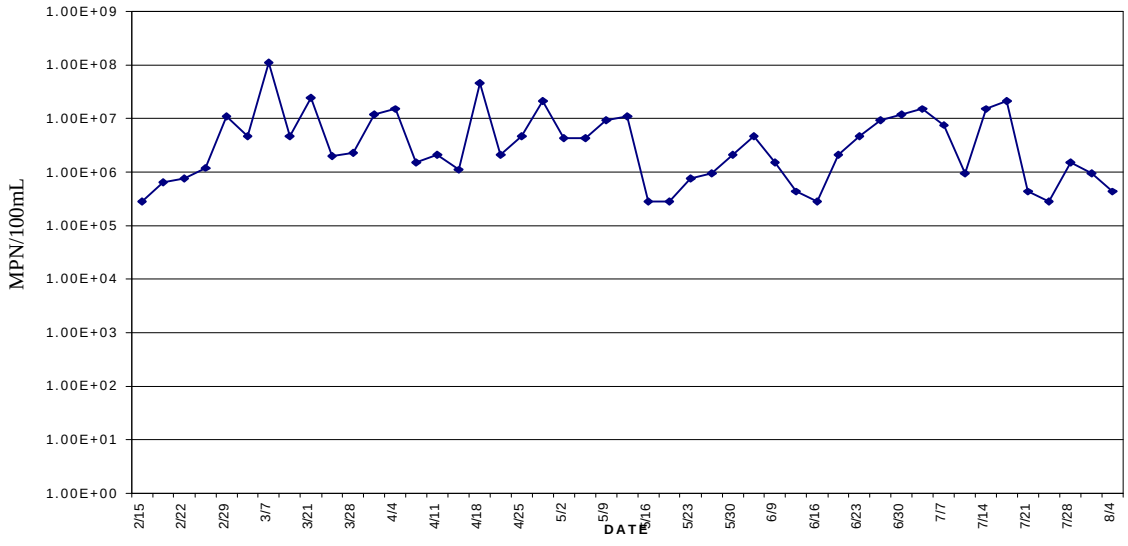

Figure A.6 Substrate - denitrifiers

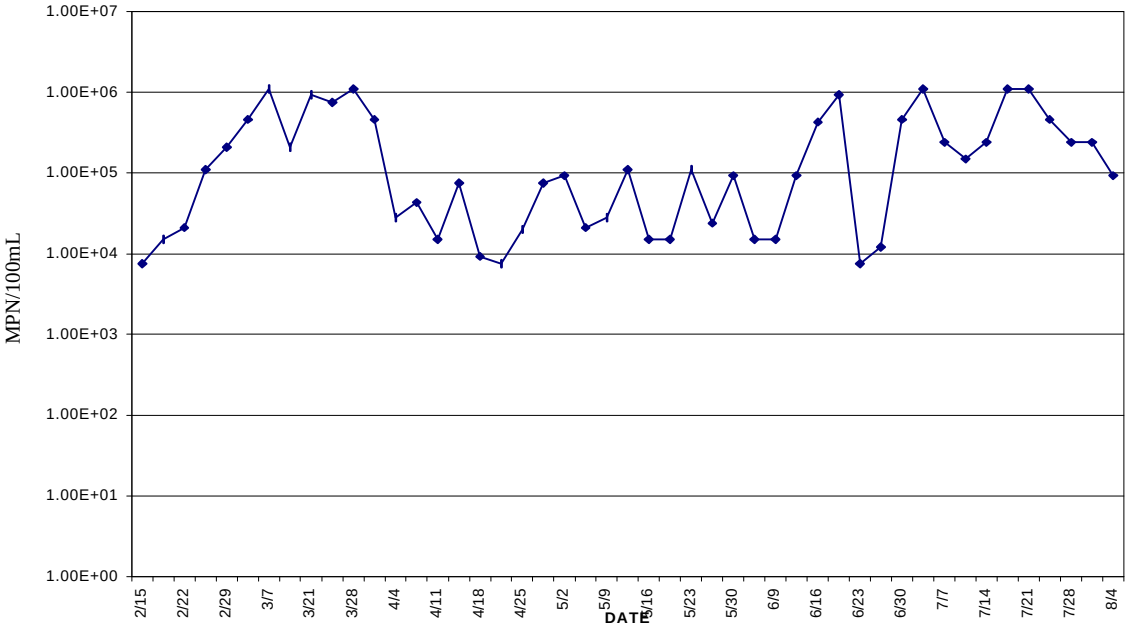

Figure A.7 Substrate – sulfate reducers

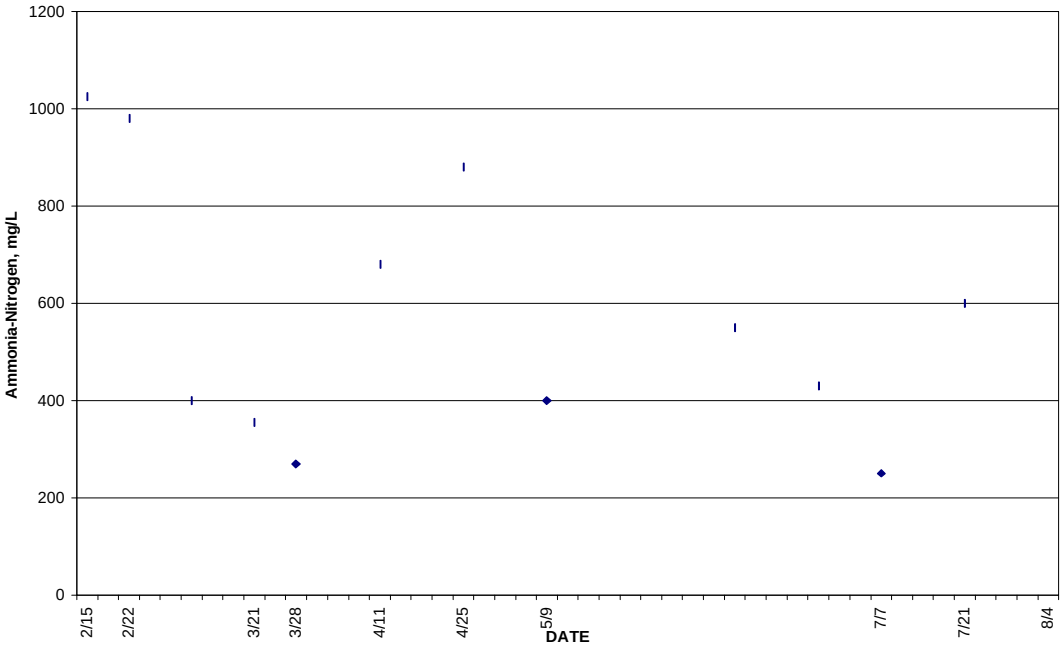

Figure A.8 Substrate – ammonia-nitrogen

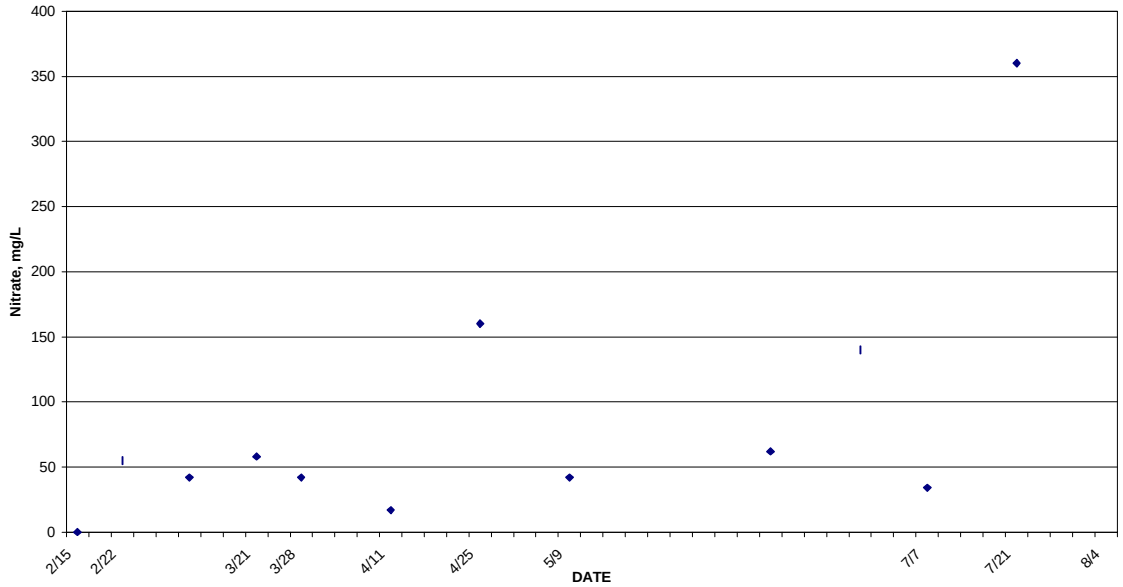

Figure A.9 Substrate – nitrate-nitrogen

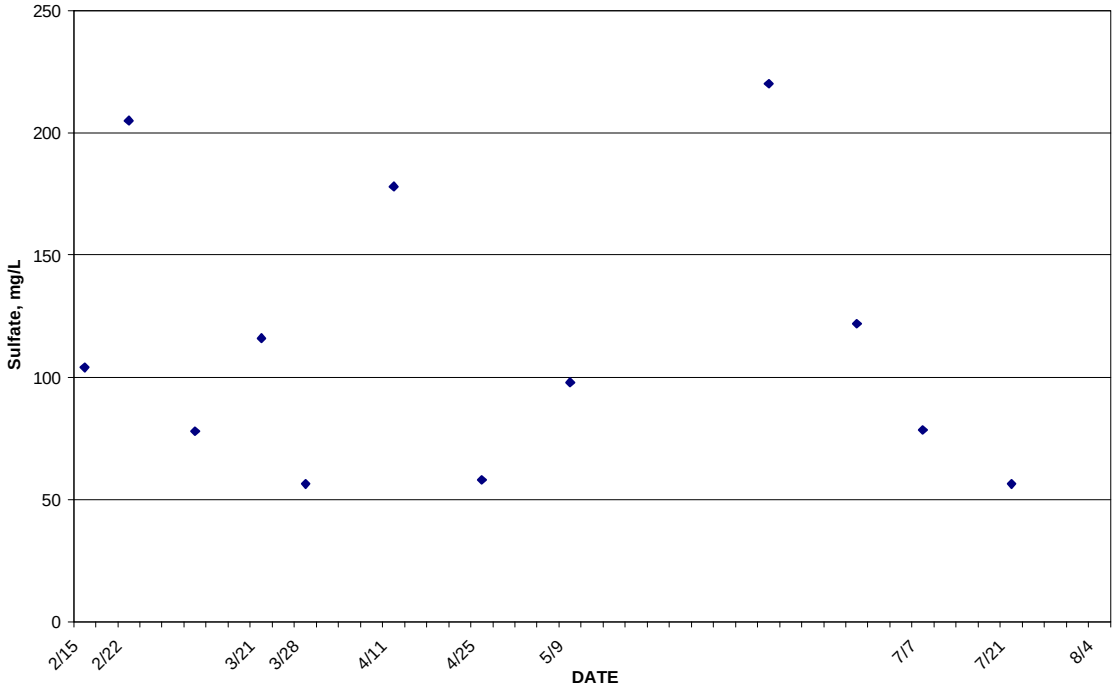

Figure A.10 Substrate - sulfate

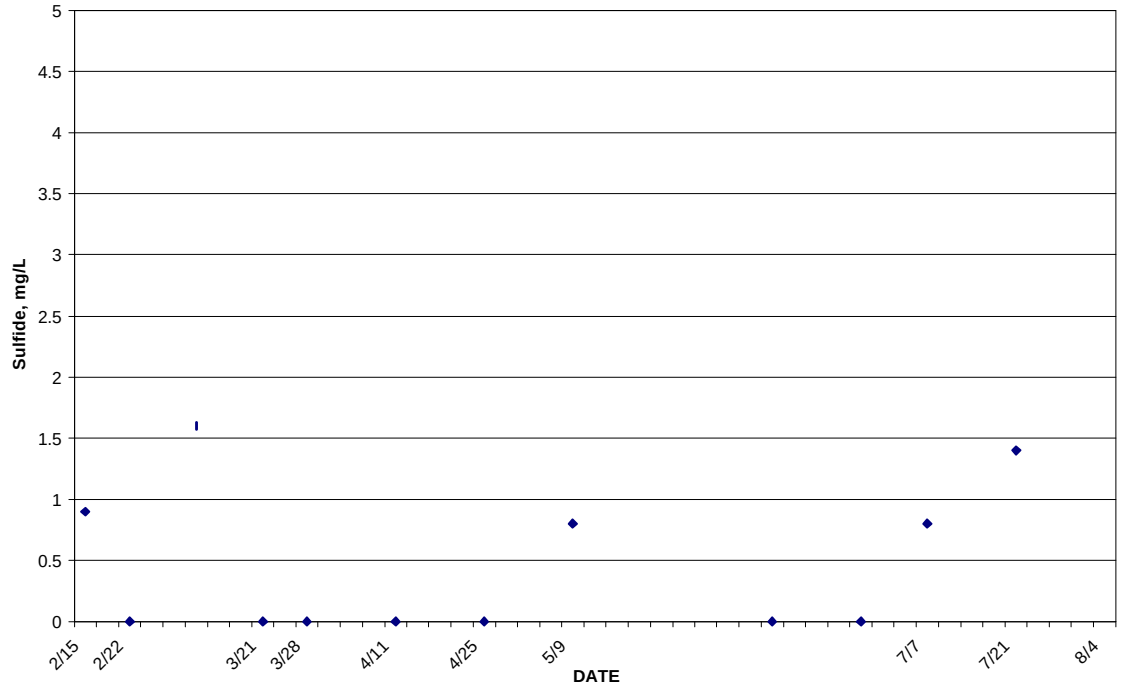

Figure A.11 Substrate - sulfide

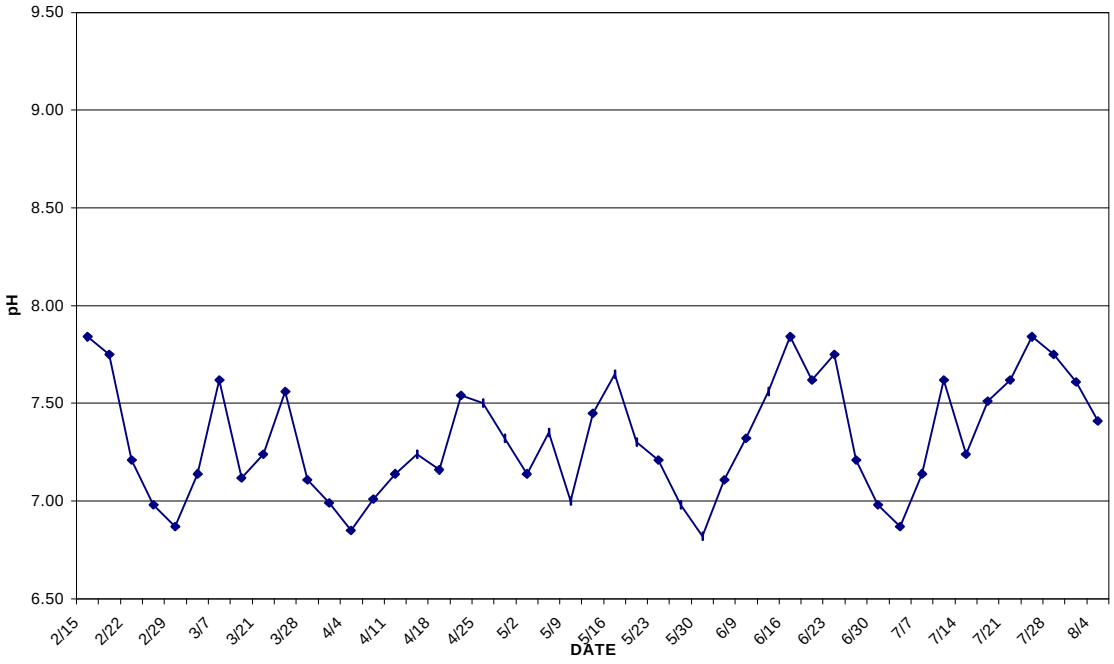

Figure A.12 Substrate - pH

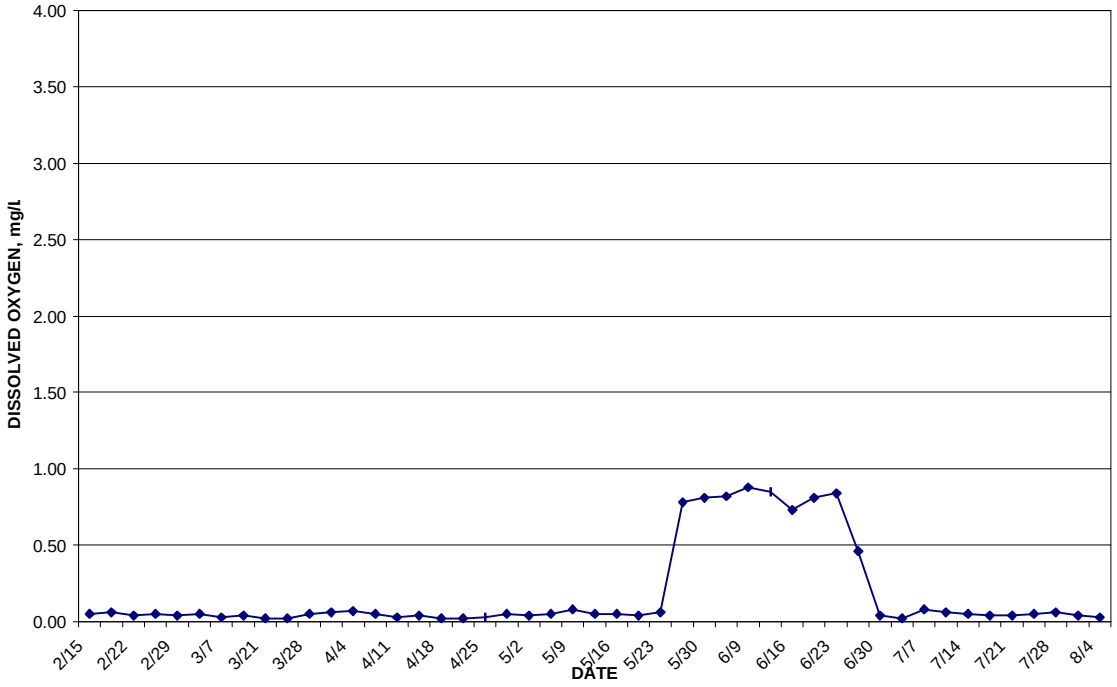

Figure A.13 Substrate – dissolved oxygen

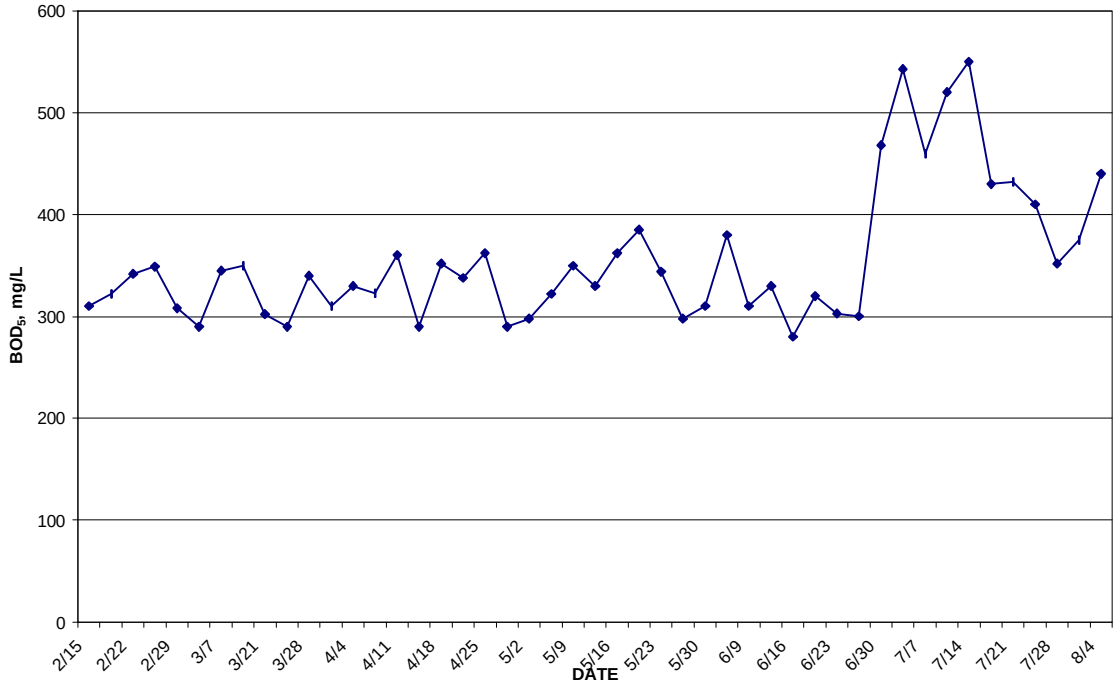

Figure A.14 Substrate - BOD

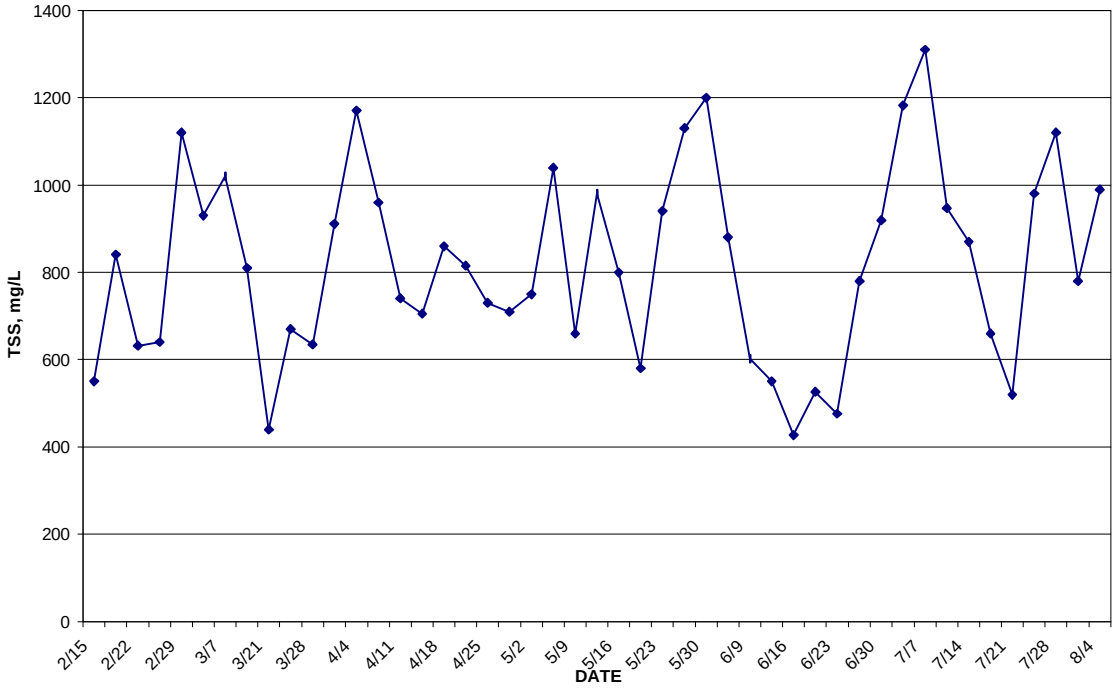

Figure A.15 Substrate – total suspended solids

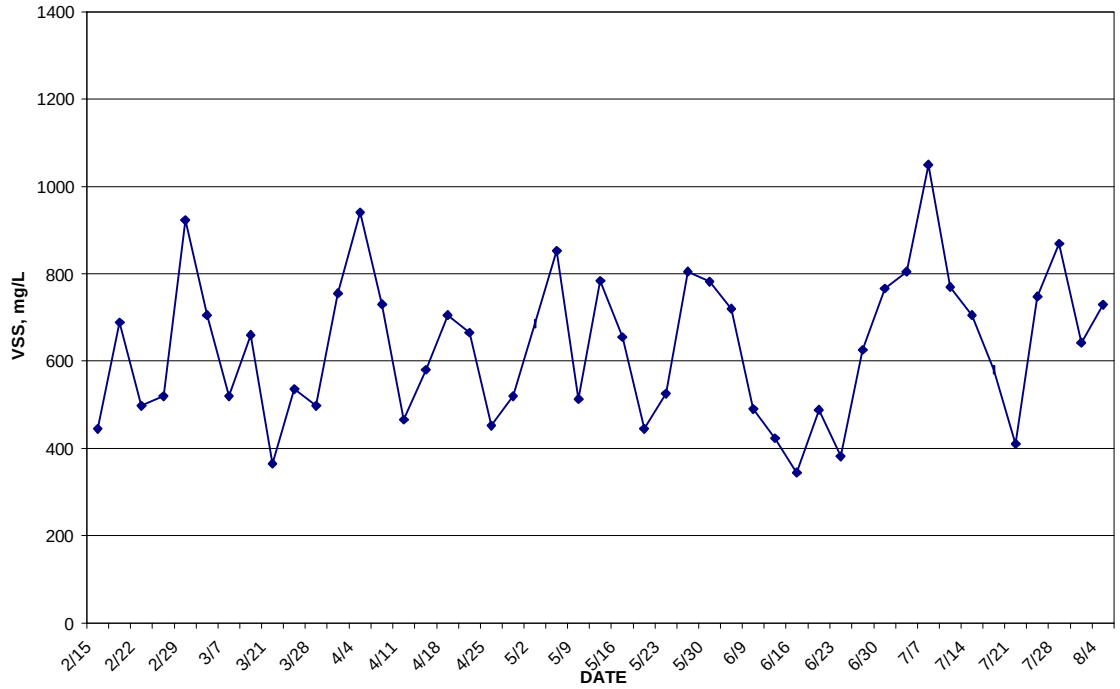

Figure A.16 Substrate – volatile suspended solids

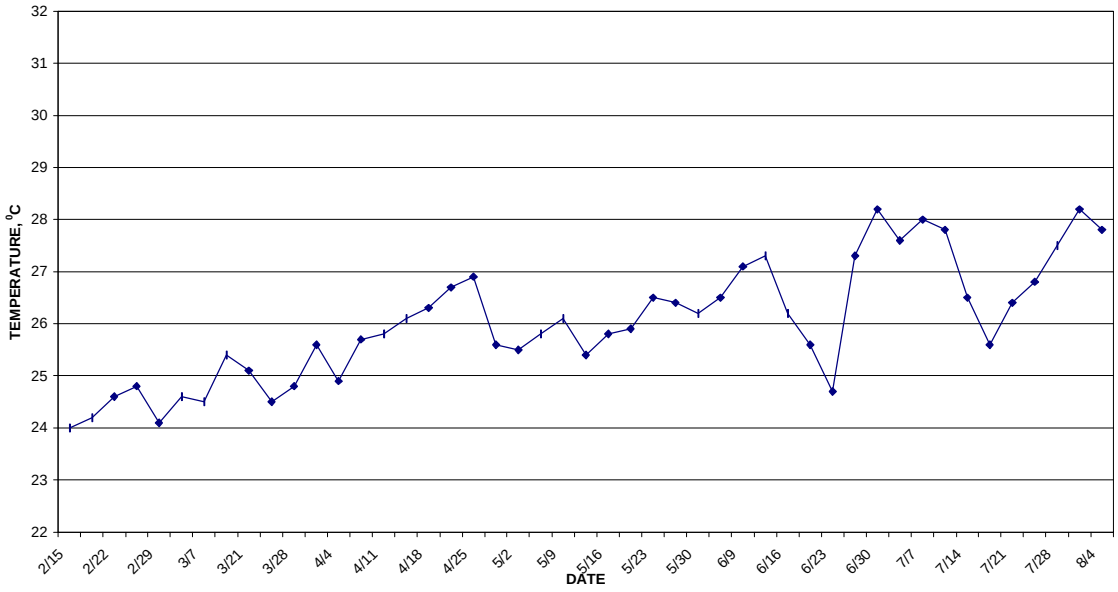

Figure A.17 Substrate - temperature

APPENDIX B

PILOT REACTOR - BOTTOM DATA ANALYSIS

Table B.1 Pilot Reactor – Bottom Data Analysis

PARAMETER	3/16	3/19	3/23	3/26	3/30	4/2	4/7	4/13	4/21	4/24	4/29	5/1	5/4	4/7	4/11	4/14	4/18	4/21	4/25	4/28	5/2	5/5	5/9
REACTOR BOTTOM-TOTAL PLATE COUNT-AEROBIC	2.00E+06	2.80E+06	2.20E+06	2.00E+06	1.20E+06	1.00E+06	6.00E+07	1.04E+06	4.80E+07	7.00E+07	2.20E+06	1.20E+06	5.60E+07	5.00E+07	1.10E+06	1.30E+06	1.10E+06	2.30E+06	1.80E+06	1.04E+06	9.80E+07	1.20E+06	5.50E+07
REACTOR BOTTOM-TOTAL PLATE COUNT-ANAEROBIC	3.00E+06	1.00E+06	9.80E+07	7.80E+07	3.20E+06	1.00E+06	7.80E+06	9.20E+07	5.00E+07	5.60E+07	9.80E+07	4.00E+07	4.00E+07	3.20E+07	7.80E+07	5.60E+07	4.00E+07	3.20E+07	4.80E+07	6.20E+07	4.30E+07	7.50E+07	4.40E+07
REACTOR BOTTOM-HYDROGEN SULFIDE PRODUCERS	1.50E+06	9.80E+04	4.30E+04	3.90E+04	4.60E+04	4.60E+05	1.50E+05	3.20E+04	4.30E+04	3.50E+04	4.30E+04	4.60E+05	5.00E+05	4.00E+05	4.80E+05	5.00E+05	4.30E+05	5.20E+05	4.00E+04	3.70E+04	1.10E+06	6.00E+05	6.20E+05
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+06	4.30E+05	7.50E+05	9.30E+05	1.20E+06	9.30E+05	1.50E+05	2.10E+05	2.00E+05	2.30E+05	1.10E+05	2.40E+05	1.10E+07	4.60E+05	1.10E+05	1.10E+07	2.40E+07	2.30E+05	1.50E+05	2.40E+05	1.10E+05	1.10E+07	4.60E+07
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	1.10E+06	4.30E+05	7.50E+05	9.30E+05	1.20E+06	9.30E+05	1.50E+05	2.10E+05	2.00E+05	2.30E+05	1.10E+05	9.50E+04	1.10E+07	4.60E+05	1.10E+05	1.10E+07	2.40E+07	2.30E+05	1.50E+05	2.40E+05	1.10E+05	1.10E+07	4.60E+07
REACTOR BOTTOM-DENITRIFIERS	2.30E+06	2.40E+06	4.60E+06	1.10E+07	9.30E+06	1.20E+07	1.10E+07	2.30E+06	2.30E+06	4.60E+06	2.10E+07	1.50E+07	4.30E+07	1.50E+06	2.30E+07	1.50E+06	7.50E+06	2.40E+06	1.10E+06	2.30E+06	2.40E+06	1.50E+06	2.00E+06
REACTOR BOTTOM-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.90E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	2.30E+01	4.30E+01	7.50E+01	7.50E+01	9.30E+01	7.50E+01	9.30E+01	2.40E+02	9.30E+02	1.50E+03	9.30E+02	2.10E+03	2.40E+03	1.20E+03
REACTOR BOTTOM-AMMONIA-N	1025		960			400			310		290				275				330				310
REACTOR BOTTOM-NITRATE	0.0		66.0			42.0			23.2		19.8				8.3				18.5				4.3
REACTOR BOTTOM-SULFATE	104.0		206.0			78.0			22.0		18.0				10.0				22.0				14.9
REACTOR BOTTOM-SULFIDE	0.9		0.0			1.8			0.0		0.0				0.0				0.0				0.0
REACTOR BOTTOM-pH	7.66	7.75	7.58	7.48	7.56	7.66	7.71	7.75	7.63	7.77	8.01	7.95	7.98	8.16	8.29	8.16	8.36	8.44	8.62	8.72	8.62	8.66	8.66
REACTOR BOTTOM-DISSOLVED OXYGEN	0.06	0.05	0.06	0.06	0.09	0.06	0.16	0.18	0.07	0.09	0.11	0.07	0.04	0.06	0.09	0.02	0.19	0.21	0.32	0.16	0.24	0.30	0.31
REACTOR TEMPERATURE	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	27.2	26.6	26.6	27.1	26.9	26.6	27.1	27.4	27.6	27.4	27.2	26.9
REACTOR BOTTOM-BOD	114	116	120	124	110	100	112	120	98	107	96	72	88	106	112	70	96	88	110	86	100	112	70
REACTOR BOTTOM-TSS	466	486	520	570	602	423	474	492	564	523	556	754	621	553	589	621	657	519	536	661	522	566	627
REACTOR BOTTOM-VSS	255	280	290	326	344	222	266	279	303	260	325	456	450	466	323	340	348	276	306	306	267	323	353

PARAMETER	5/12	5/16	5/19	5/23	5/26	5/30	6/6	6/9	6/13	6/16	6/20	6/23	6/27	6/30	7/4	7/7	7/11	7/14	7/18	7/21	7/28	7/28	8/1	8/4
REACTOR BOTTOM-TOTAL PLATE COUNT-AEROBIC	5.00E+07	9.20E+07	1.60E+06	5.50E+07	1.20E+06	1.90E+06	1.04E+06	5.60E+07	5.00E+07	9.20E+07	1.00E+06	9.80E+07	1.04E+06	1.04E+06	9.80E+07	5.50E+07	1.00E+06	1.80E+06	9.80E+07	1.12E+06	1.50E+06	9.80E+07	9.50E+07	5.50E+07
REACTOR BOTTOM-TOTAL PLATE COUNT-ANAEROBIC	4.60E+07	4.60E+07	9.80E+07	1.04E+06	3.20E+07	5.50E+07	9.00E+07	4.60E+07	4.60E+07	5.60E+07	9.00E+07	4.50E+07	4.80E+07	4.80E+07	5.50E+07	9.00E+07	1.60E+06	1.20E+06	5.50E+07	9.00E+07	3.50E+07	6.20E+07	6.80E+07	1.04E+07
REACTOR BOTTOM-HYDROGEN SULFIDE PRODUCERS	5.50E+04	3.20E+04	2.20E+04	4.80E+06	4.40E+05	5.50E+05	5.00E+06	4.00E+05	9.00E+04	4.50E+06	5.00E+06	3.20E+05	5.50E+04	2.20E+04	2.20E+04	5.50E+04	4.50E+04	2.50E+04	4.80E+04	3.20E+04	2.80E+04	3.00E+04	2.20E+04	3.20E+04
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+06	1.20E+07	9.30E+07	9.30E+07	2.10E+07	9.30E+05	1.20E+06	4.30E+05	9.50E+05	1.10E+07	1.10E+06	2.40E+05	2.10E+06	2.30E+06	4.30E+05	1.10E+06	4.60E+06	2.40E+05	4.30E+05	4.60E+06	2.10E+05	4.30E+05	9.30E+06	9.30E+05
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	4.60E+07	1.20E+07	9.30E+07	9.30E+07	2.10E+07	9.30E+05	1.20E+06	4.30E+05	9.30E+05	1.10E+07	4.60E+06	2.40E+05	2.10E+06	2.30E+06	4.30E+05	1.10E+06	4.60E+06	2.40E+05	4.30E+05	4.60E+06	2.10E+05	4.30E+05	9.30E+06	9.30E+05
REACTOR BOTTOM-DENITRIFIERS	1.50E+06	2.30E+06	1.50E+07	9.30E+06	4.60E+06	4.60E+06	2.30E+06	2.30E+06	7.50E+06	4.60E+07	7.50E+06	1.10E+05	2.00E+05	2.10E+05	2.30E+05	1.10E+07	4.50E+07	1.10E+07	4.60E+07	2.30E+06	2.30E+06	9.30E+06	2.40E+06	1.10E+06
REACTOR BOTTOM-SULFATE REDUCERS	2.40E+03	4.60E+03	2.40E+03	4.60E+03	1.10E+04	1.10E+04	2.40E+03	9.30E+03	4.60E+03	7.50E+03	9.30E+03	1.10E+04	2.10E+04	2.10E+04	2.00E+04	1.50E+04	9.30E+03	7.50E+03	9.30E+03	7.50E+03	2.40E+02	4.60E+02	1.10E+03	2.10E+02
AMMONIA-N									410				334			310				360				
NITRATE									1.8				2.3			0.0				1.1				
SULFATE									5.5				14.3			2.3				22.6				
SULFIDE									0.0				0.0			0.0				0.0				
REACTOR BOTTOM-pH	8.44	8.43	8.66	8.66	8.67	8.66	8.66	8.66	8.62	8.66	8.38	8.62	8.26	8.47	8.64	8.99	8.99	8.99	8.84	8.86	8.92	8.66	8.96	8.92
REACTOR BOTTOM-DISSOLVED OXYGEN	0.25	0.21	0.18	0.14	0.09	0.09	0.13	0.21	0.15	0.18	0.29	0.22	0.43	0.61	0.78	0.29	0.68	0.64	0.32	0.66	0.55	0.46	0.41	0.23
REACTOR TEMPERATURE	27.5	27.9	27.6	27.6	28	27.6	27.5	28.4	28.5	29.4	29.5	30.2	31.2	30	30	30	30	29.5	29.8	29.4	30	30	30.1	29.8
REACTOR BOTTOM-BOD	96	112	120	66	76	66	76	62	76	56	66	61	80	94	109	96	150	132	84	87	78	70	114	96
REACTOR BOTTOM-TSS	664	673	626	550	466	490	551	562	580	669	648	666	626	664	589	632	631	521	660	652	560	548	660	576
REACTOR BOTTOM-VSS	366	325	266	296	275	256	303	312	321	367	360	376	266	466	323	302	346	274	366	360	335	260	304	316

Table B.2 Reactor Bottom – Characterization Plots

FIGURES B.1 THROUGH B.17		
B.1	REACTOR BOTTOM	TOTAL PLATE COUNT - AEROBIC
B.2	REACTOR BOTTOM	TOTAL PLATE COUNT - ANAEROBIC
B.3	REACTOR BOTTOM	HYDROGEN-SULFIDE PRODUCERS
B.4	REACTOR BOTTOM	CARBOHYDRATE-UTILIZERS - ACID PRODUCERS
B.5	REACTOR BOTTOM	CARBOHYDRATE-UTILIZERS - GAS PRODUCERS
B.6	REACTOR BOTTOM	DENITRIFIERS
B.7	REACTOR BOTTOM	SULFATE REDUCERS
B.8	REACTOR BOTTOM	AMMONIA-NITROGEN
B.9	REACTOR BOTTOM	NITRATE-NITROGEN
B.10	REACTOR BOTTOM	SULFATE
B.11	REACTOR BOTTOM	SULFITE
B.12	REACTOR BOTTOM	pH
B.13	REACTOR BOTTOM	DISSOLVED OXYGEN
B.14	REACTOR BOTTOM	BOD
B.15	REACTOR BOTTOM	TOTAL SUSPENDED SOLIDS
B.16	REACTOR BOTTOM	VOLATILE SUSPENDED SOLIDS
B.17	REACTOR BOTTOM	TEMPERATURE

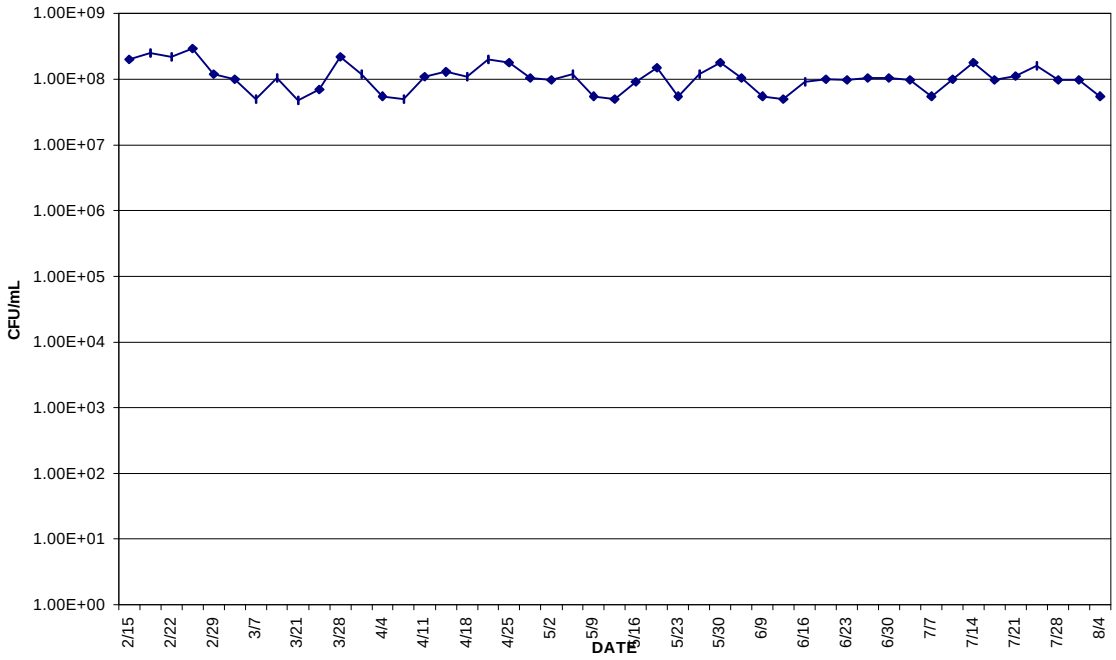

Figure B.1 Reactor bottom – total plate count - aerobic

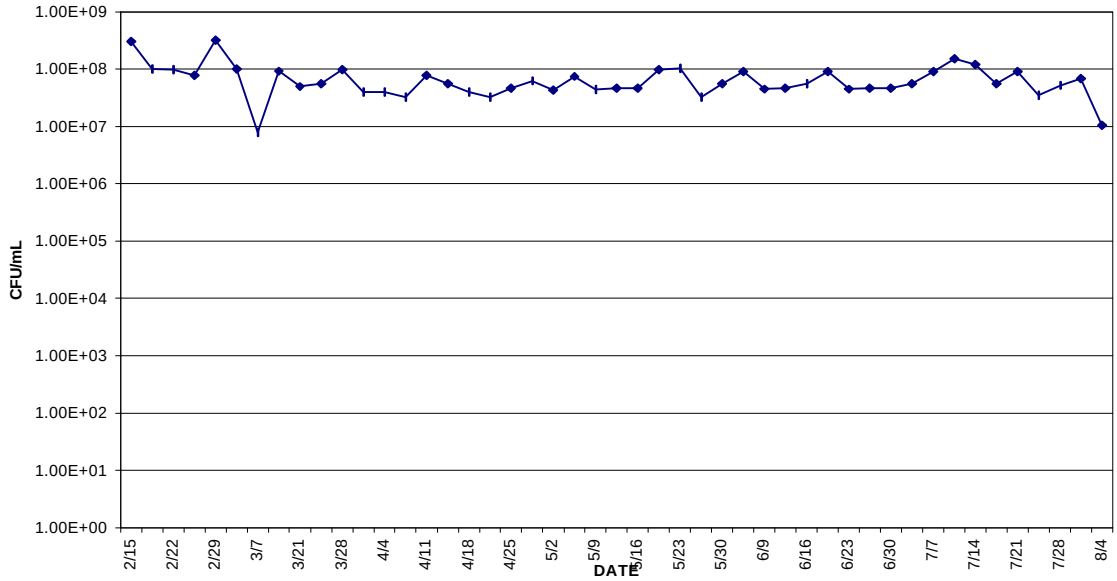

Figure B.2 Reactor bottom – total plate count - anaerobic

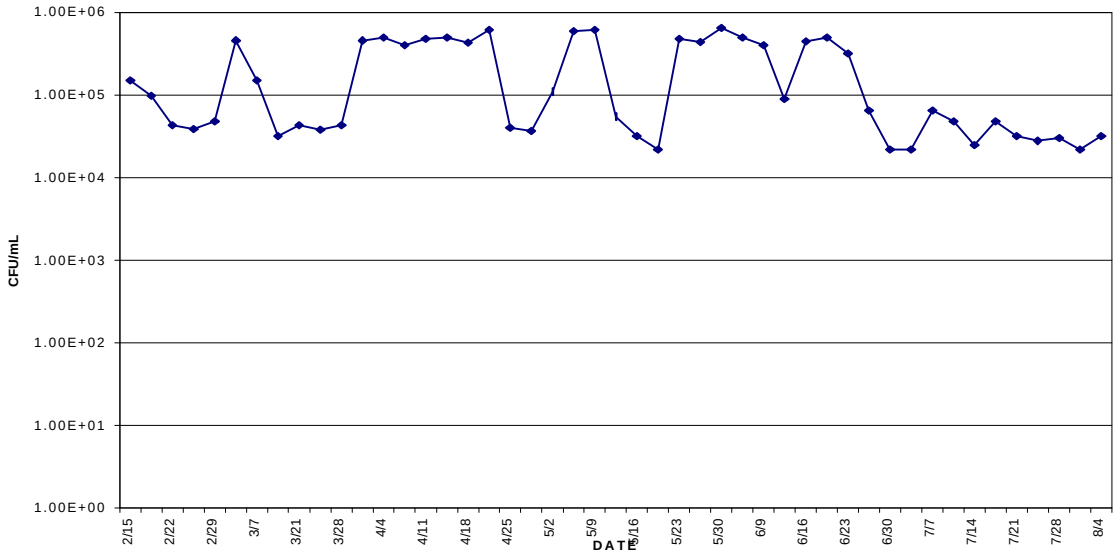

Figure B.3 Reactor bottom – hydrogen-sulfide producers

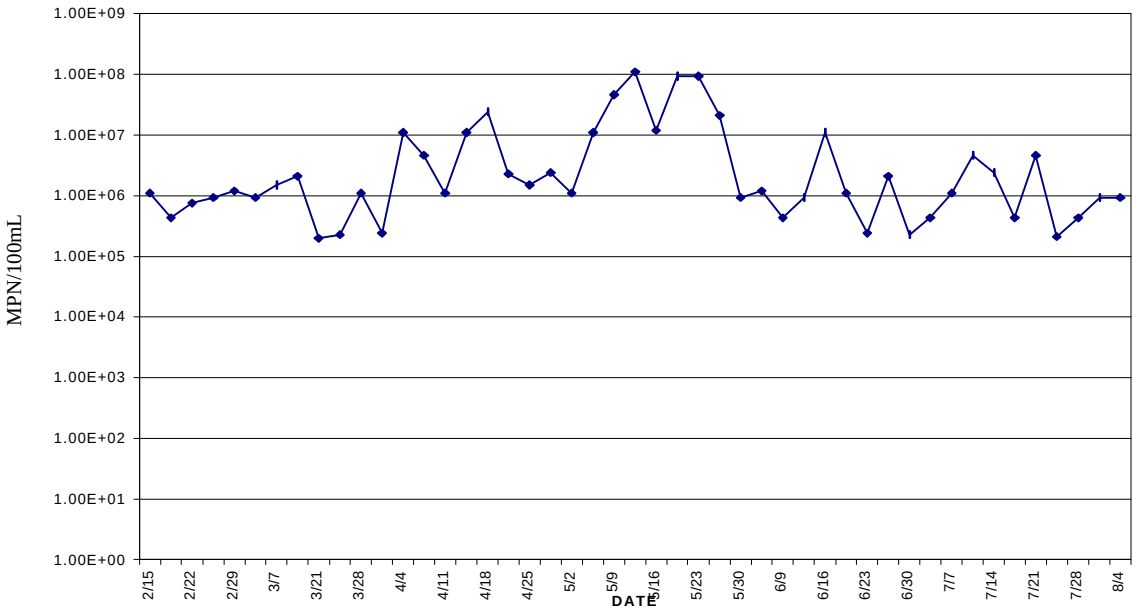

Figure B.4 Reactor bottom – carbohydrate-utilizers – acid-producers

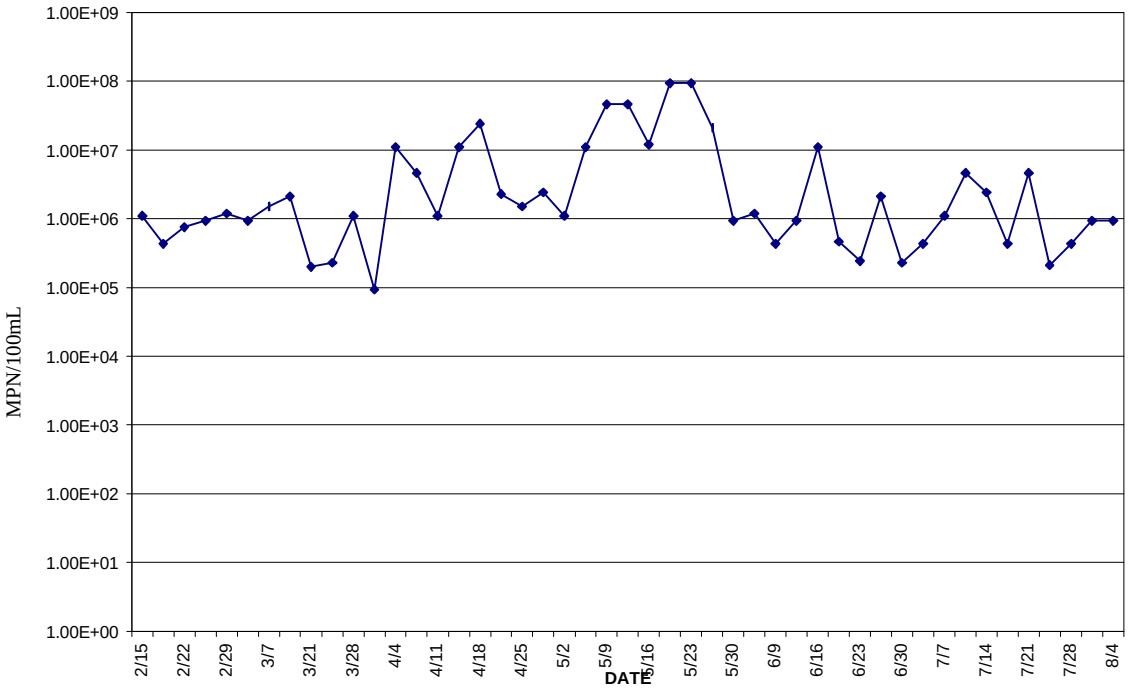

Figure B.5 Reactor bottom – carbohydrate-utilizers – gas producers

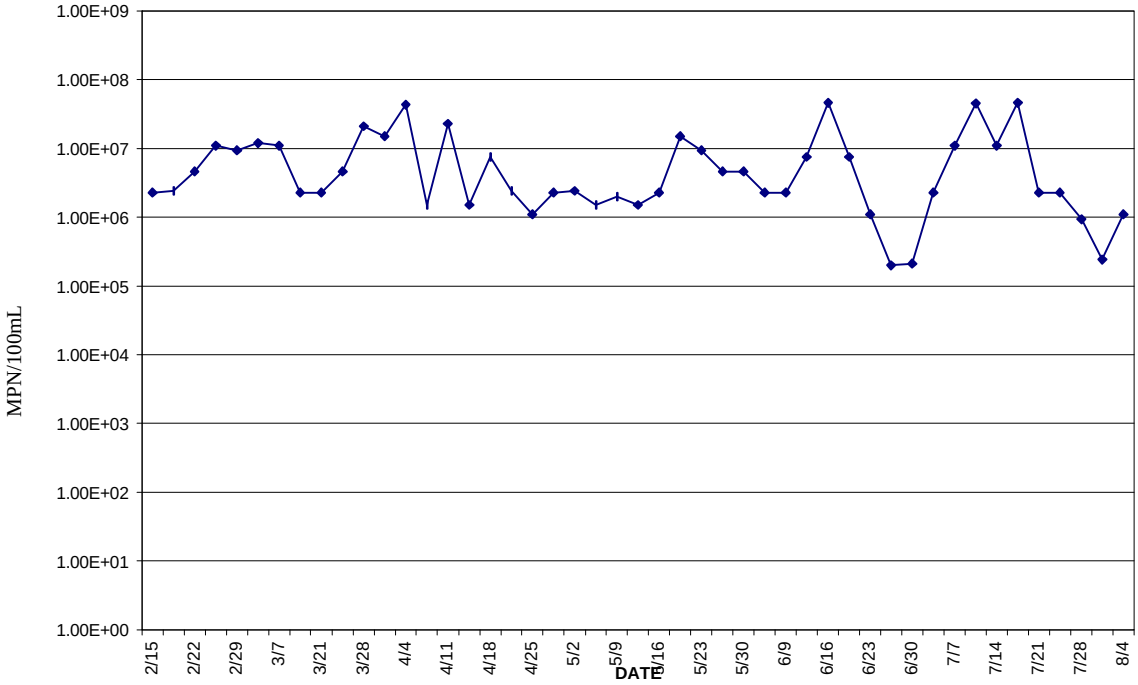

Figure B.6 Reactor bottom - denitrifiers

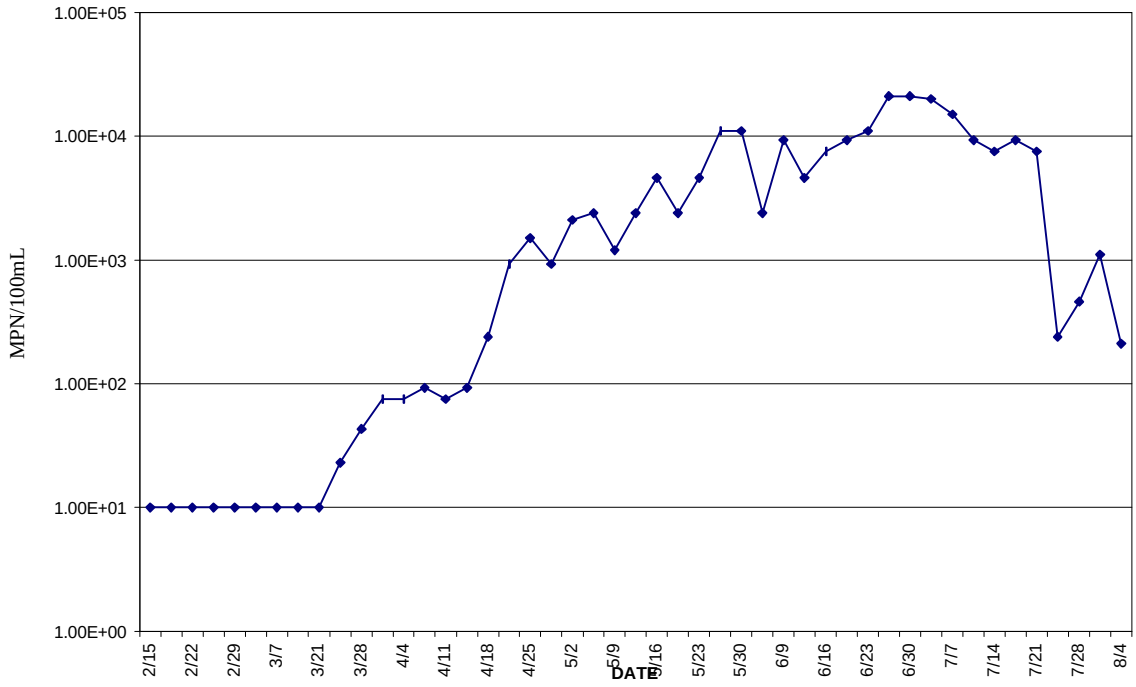

Figure B.7 Reactor bottom – sulfate-reducers

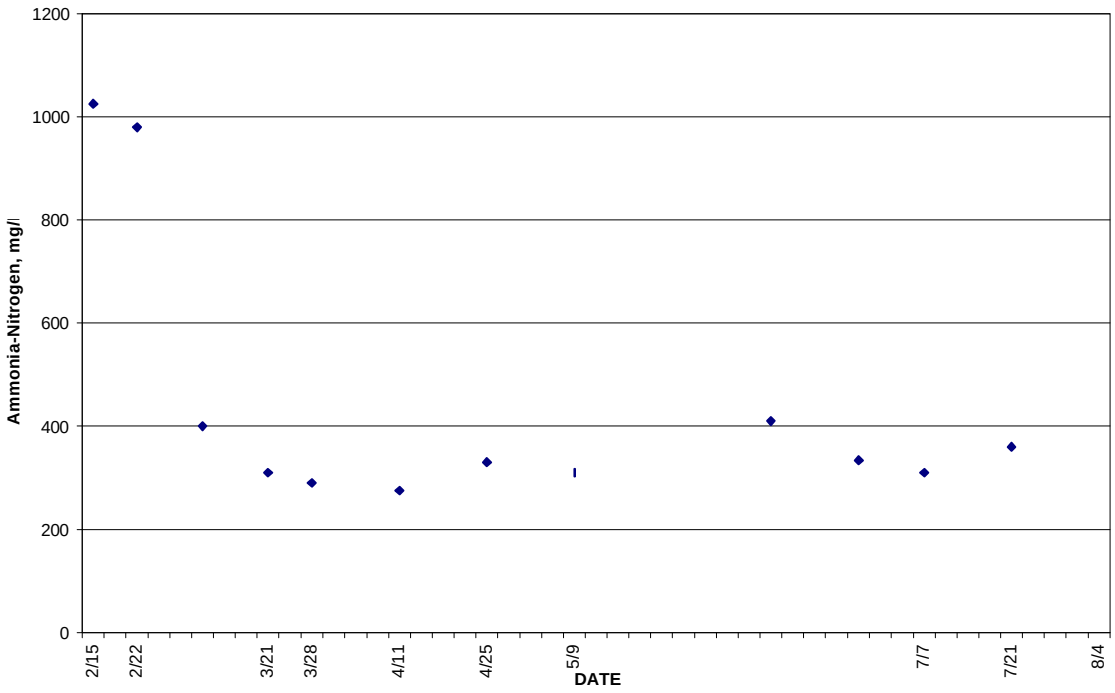

Figure B.8 Reactor bottom – ammonia-nitrogen

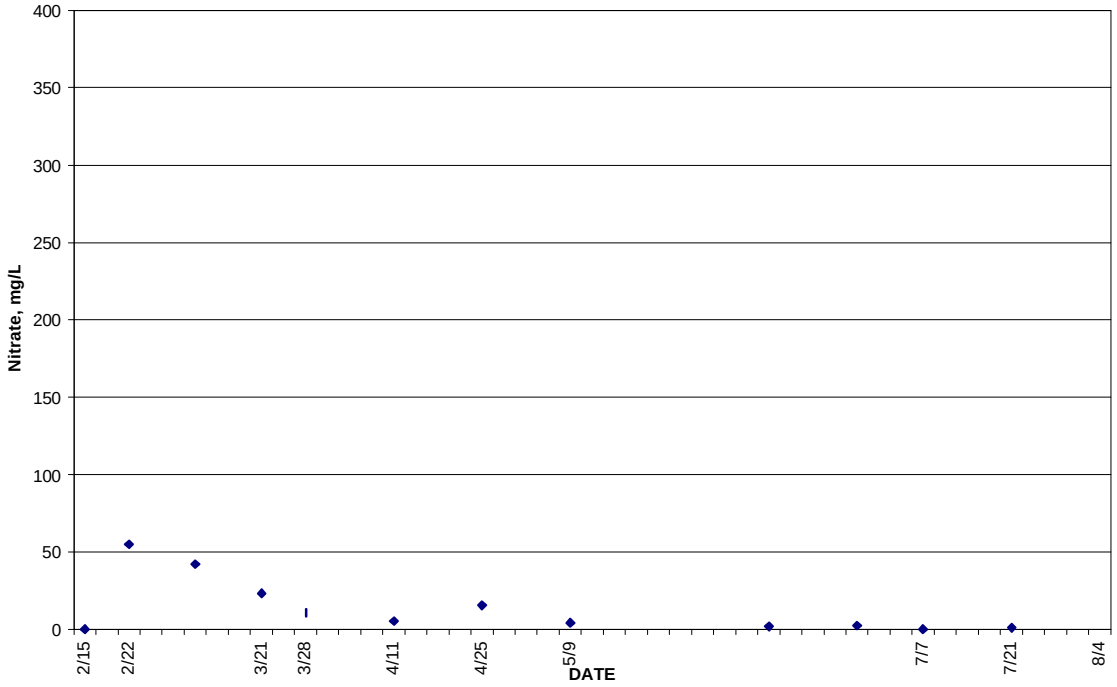

Figure B.9 Reactor bottom – nitrate-nitrogen

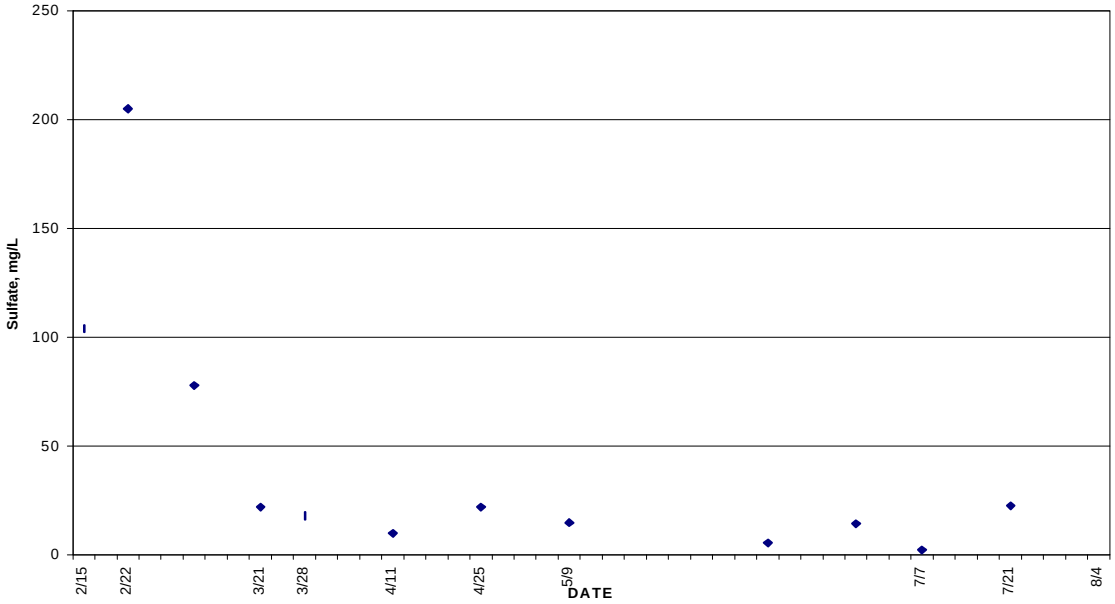

Figure B.10 Reactor bottom - sulfate

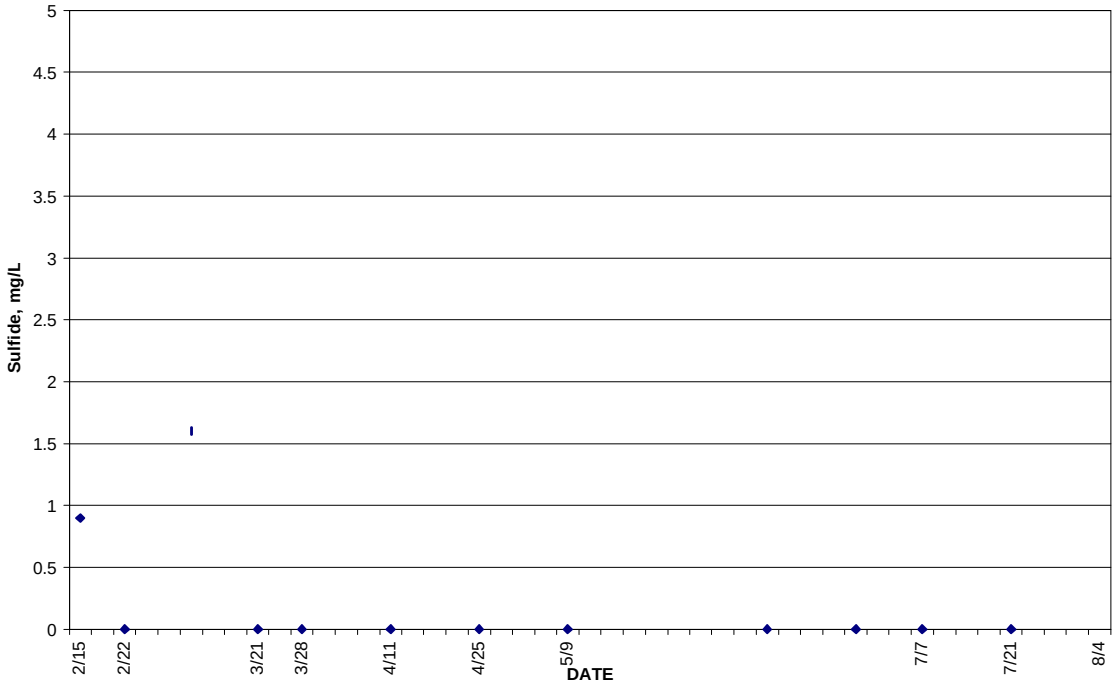

Figure B.11 Reactor bottom - sulfide

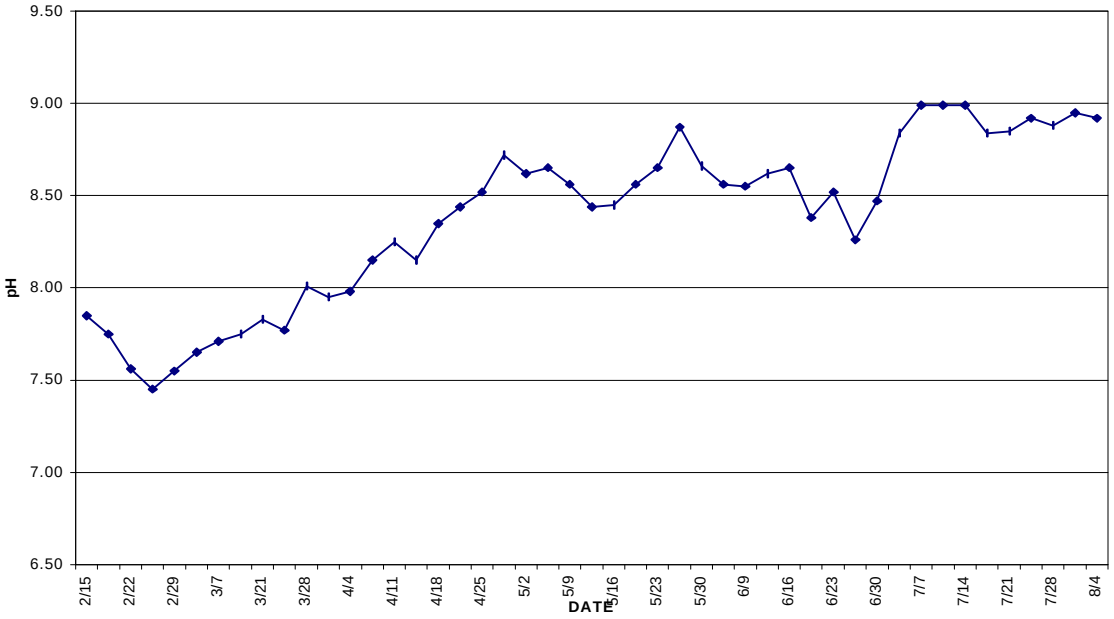

Figure B.12 Reactor bottom - pH

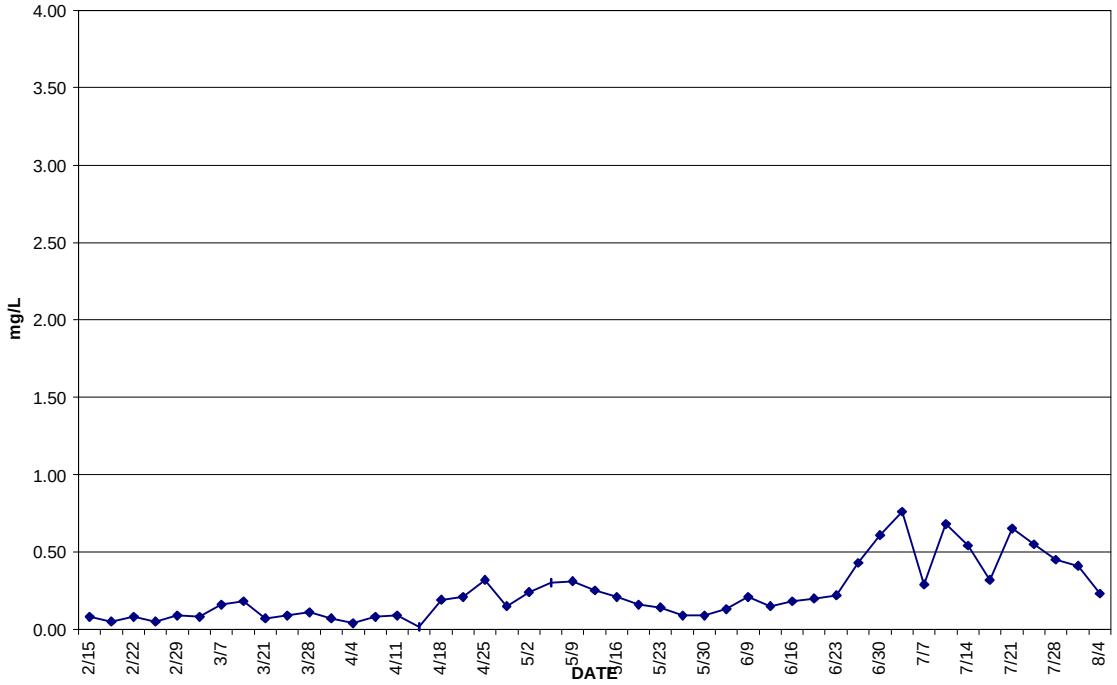

Figure B.13 Reactor bottom – dissolved oxygen

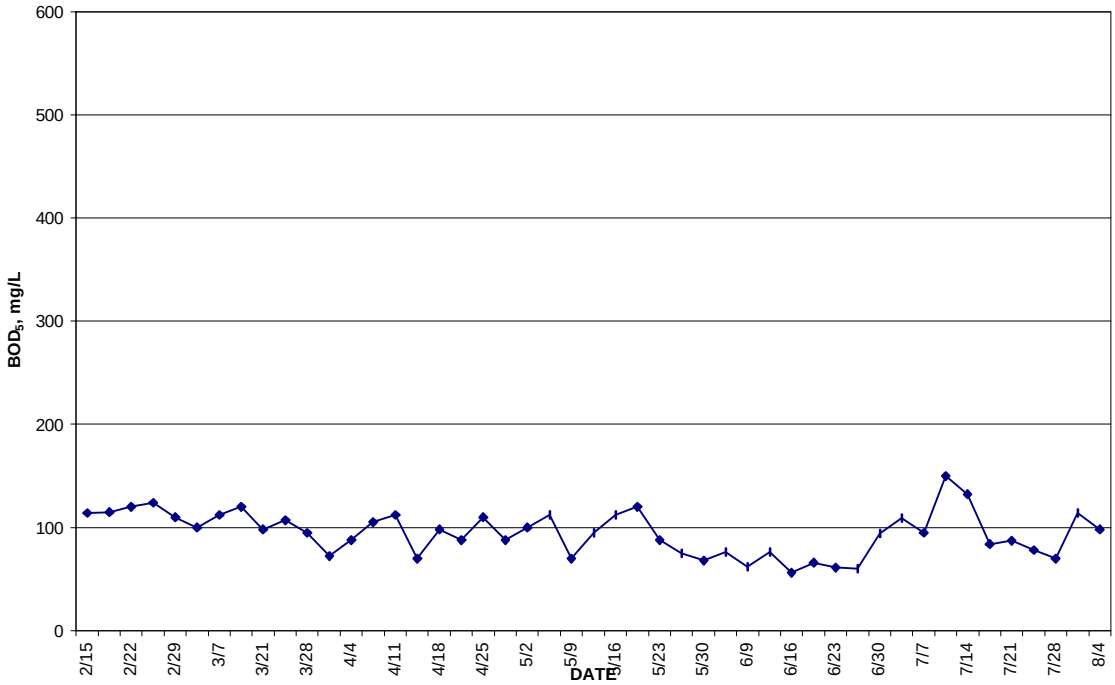

Figure B.14 Reactor bottom - BOD

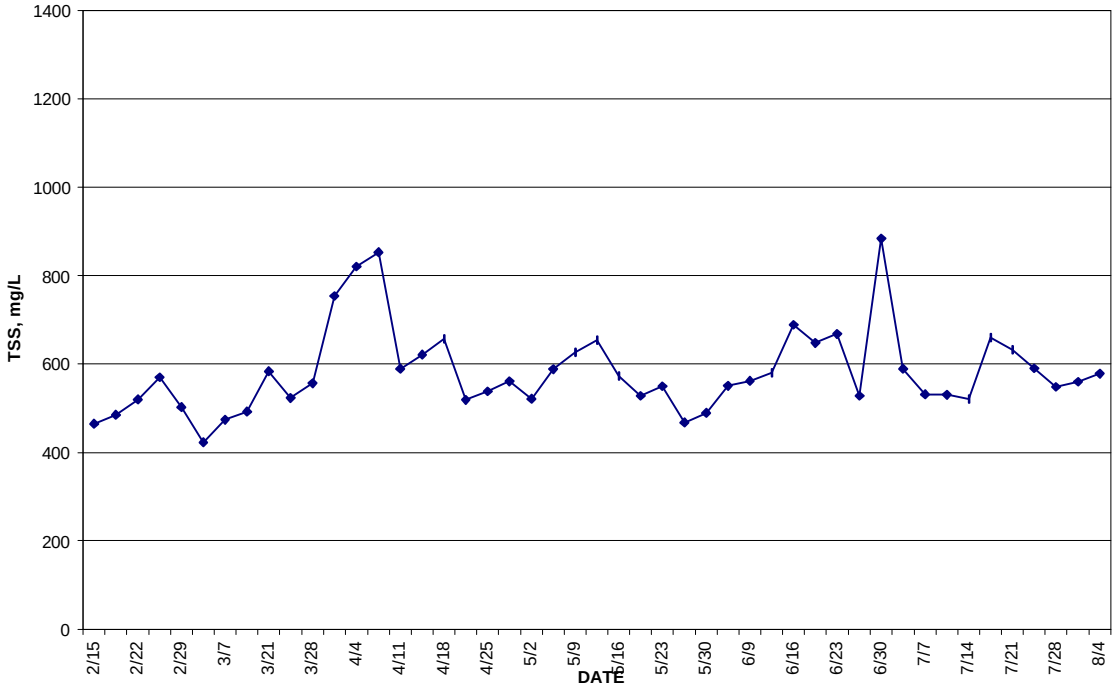

Figure B.15 Reactor bottom – total suspended solids

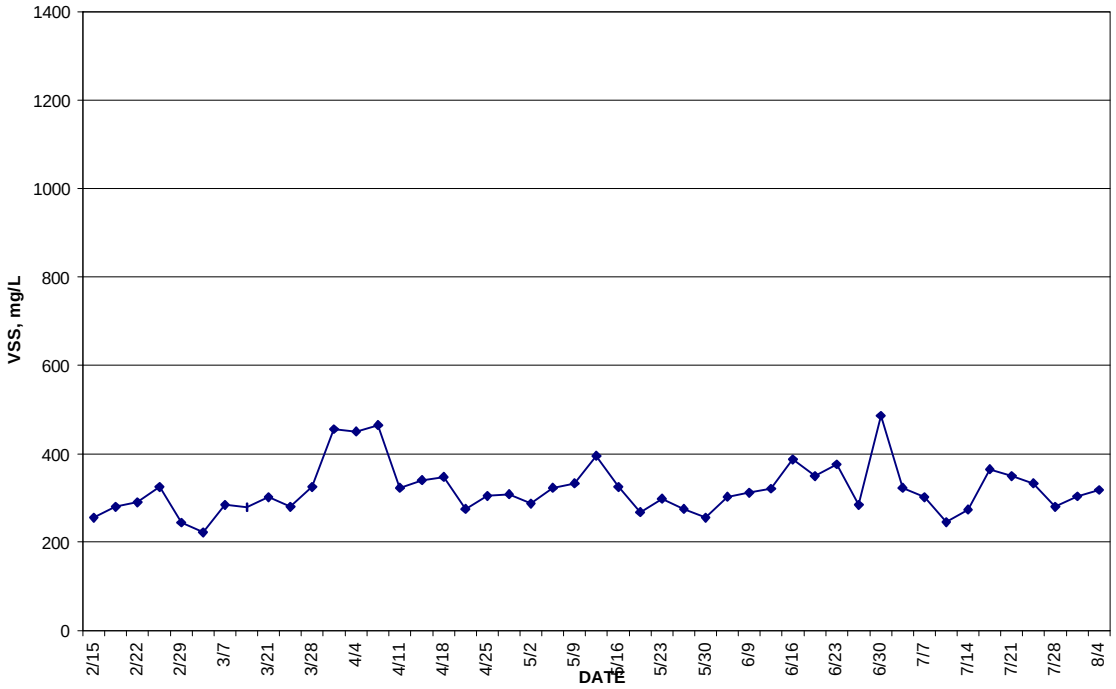

Figure B.16 Reactor bottom – volatile suspended solids

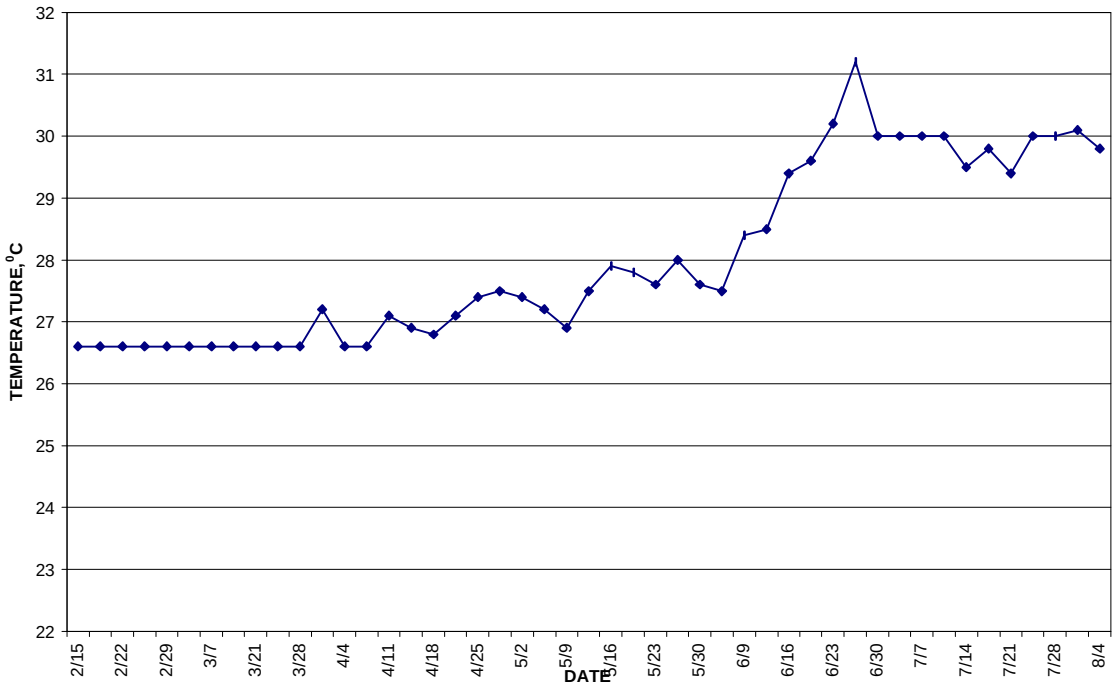

Figure B.17 Reactor bottom - temperature

APPENDIX C

PILOT REACTOR - TOP DATA ANALYSIS

Table C.1 Pilot Reactor – Top Data Analysis

PARAMETER	3/16	3/18	3/22	3/26	3/29	4/3	4/7	4/17	4/21	4/24	4/28	4/30	5/1	5/4	5/7	5/11	5/14	5/18	5/21	5/25	5/28	6/2	6/6	6/9
REACTOR TOP-TOTAL PLATE COUNT-AEROBIC	2.10E+08	2.60E+08	2.20E+08	2.80E+08	1.30E+08	1.00E+08	9.00E+07	1.10E+08	9.80E+07	1.07E+08	2.20E+08	1.22E+08	9.80E+07	7.80E+07	1.20E+08	1.30E+08	1.20E+08	2.00E+08	1.80E+08	1.09E+08	1.04E+08	1.20E+08	9.60E+07	
REACTOR TOP-TOTAL PLATE COUNT-ANAEROBIC	3.00E+07	3.20E+07	3.40E+07	6.80E+07	1.10E+08	9.20E+07	8.70E+07	8.90E+07	1.00E+08	1.02E+08	6.20E+07	3.40E+07	2.20E+07	8.80E+06	4.20E+08	1.00E+08	9.20E+06	6.20E+06	3.80E+06	2.20E+05	3.20E+06	1.00E+06	4.80E+04	
REACTOR TOP-HYDROGEN SULFIDE PRODUCERS	1.50E+05	9.60E+04	4.30E+04	3.80E+04	4.60E+04	4.60E+05	1.50E+05	3.20E+04	4.30E+04	3.80E+04	4.30E+04	4.60E+05	5.00E+05	8.00E+04	2.20E+03	1.00E+01	1.00E+01	1.00E+01	1.00E+01	6.00E+04	1.00E+01	1.00E+01	1.00E+01	
REACTOR TOP-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.30E+05	2.10E+06	2.80E+05	1.50E+06	4.60E+06	1.20E+06	1.50E+07	1.10E+07	2.00E+08	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07	
REACTOR TOP-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.30E+05	2.10E+06	2.80E+05	1.50E+06	4.60E+06	1.20E+06	1.50E+07	1.10E+07	2.00E+08	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07	
REACTOR TOP-DENITRIFIERS	4.30E+06	7.50E+06	4.30E+06	2.30E+06	2.10E+06	2.80E+06	4.30E+06	4.30E+06	2.30E+06	9.30E+06	7.50E+06	4.30E+06	7.80E+06	1.10E+07	2.10E+07	2.80E+07	1.10E+07	7.50E+06	2.30E+06	9.30E+06	2.80E+06	2.80E+07	7.50E+06	
REACTOR TOP-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	9.00E+02	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR TOP-AMMONIA-N	1025		980			400			310		290					260				300				266
REACTOR TOP-NITRATE	0.0		55.0			42.0			23.2		10.8					1.2				2.6				0.9
REACTOR TOP-SULFATE	154.0		265.0			78.0			22.0		0.6					0.0				0.0				0.0
REACTOR TOP-SULFIDE	0.9		0.0			1.6			0.0		0.0					0.0				0.0				0.0
REACTOR TOP-pH	7.85	7.75	7.86	7.45	7.55	7.56	7.71	7.35	7.63	7.77	8.01	7.96	7.96	8.02	8.2	8.25	8.45	8.42	8.55	8.66	8.65	8.72	8.56	
REACTOR TOP-DISSOLVED OXYGEN	0.06	0.05	0.06	0.05	0.09	0.06	0.16	0.18	2.29	2.2	2.29	2.23	2.1	2.3	2	2	2.32	2.21	2	2.2	2	2.1	2.31	
REACTOR TOP-TEMPERATURE	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	27.2	26.6	26.6	27.1	26.9	26.6	27.1	27.4	27.5	27.4	27.2	26.9	
REACTOR TOP-BOD	114	116	120	124	110	100	112	120	98	95	74	82	88	72	74	56	78	75	72	85	78	62	84	
REACTOR TOP-TSS	738	486	478	562	476	406	488	480	564	550	588	741	623	628	762	705	691	633	617	562	656	623	584	
REACTOR TOP-VSS	396	218	229	265	255	252	245	229	278	274	248	390	400	314	364	352	333	338	308	332	328	291	305	

PARAMETER	6/12	6/16	6/19	6/23	6/26	6/30	7/3	7/9	7/13	7/16	7/20	7/23	7/27	7/30	7/4	7/7	7/11	7/14	7/18	7/21	7/25	7/28	8/1	8/4
REACTOR TOP-TOTAL PLATE COUNT-AEROBIC	1.10E+08	1.00E+08	1.53E+08	7.50E+07	2.00E+08	2.20E+08	1.08E+08	8.80E+07	8.90E+07	1.02E+08	1.08E+08	1.10E+08	1.12E+08	1.12E+08	1.12E+08	8.80E+07	1.08E+08	1.80E+08	1.12E+08	1.14E+08	1.90E+08	7.20E+07	9.20E+07	7.40E+07
REACTOR TOP-TOTAL PLATE COUNT-ANAEROBIC	2.20E+05	4.50E+05	3.20E+05	1.04E+05	9.80E+04	8.60E+04	1.02E+06	2.20E+05	2.20E+05	3.80E+05	3.20E+05	7.20E+04	6.80E+04	2.20E+05	2.00E+05	1.00E+05	1.08E+05	6.20E+03	2.20E+05	8.20E+05	6.80E+06	6.80E+05	2.20E+05	2.02E+05
REACTOR TOP-HYDROGEN SULFIDE PRODUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR TOP-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.80E+07	2.30E+07	9.30E+06	2.00E+06	9.30E+05	1.20E+06	2.80E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.80E+06	1.80E+06	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+06	7.50E+06	1.20E+06
REACTOR TOP-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.80E+07	2.30E+07	9.30E+06	2.00E+06	9.30E+05	1.20E+06	2.80E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.80E+06	1.80E+06	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+06	7.50E+06	1.20E+06
REACTOR TOP-DENITRIFIERS	7.50E+06	2.40E+07	1.20E+07	2.40E+07	1.10E+07	7.50E+06	2.30E+06	2.80E+06	1.10E+06	1.20E+06	9.30E+05	4.60E+06	1.10E+07	2.40E+06	6.30E+04	1.10E+06	2.10E+05	2.40E+06	7.50E+05	2.40E+05	4.80E+06	2.40E+06	1.10E+06	9.30E+05
REACTOR TOP-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR TOP-AMMONIA-N									360				280			255				330				
REACTOR TOP-NITRATE									0.0				0.3			0.0				0.3				
REACTOR TOP-SULFATE									0.0				0.0			0.0				0.0				
REACTOR TOP-SULFIDE									0.0				0.0			0.0				0.0				
REACTOR TOP-pH	8.65	8.76	8.8	8.72	8.6	8.62	8.65	8.62	8.65	8.65	8.75	8.72	8.32	8.66	8.78	9.16	9.13	9.15	9.05	9.1	9.12	9.02	9.05	9.15
REACTOR TOP-DISSOLVED OXYGEN	2.2	2.2	2.1	1.98	2	2.35	2.21	2.3	2.2	1.98	2.2	2.3	2.08	3.12	3.18	3.46	3.56	3.2	2.7	2.5	2.28	2.02	2.13	2
REACTOR TOP-TEMPERATURE	27.5	27.9	27.8	27.6	28	27.6	27.5	28.4	28.5	29.4	29.6	30.2	31.2	30	30	30	30	29.5	29.8	29.4	30	30	30.1	29.8
REACTOR TOP-BOD	78	95	112	74	65	62	68	50	64	60	55	58	45	62	74	66	104	68	62	75	62	60	75	75
REACTOR TOP-TSS	625	654	814	905	869	726	621	667	668	751	866	857	685	928	748	647	647	685	659	723	766	656	675	756
REACTOR TOP-VSS	321	338	410	456	434	336	315	328	305	366	422	411	516	284	258	331	219	250	332	336	373	438	473	387

Table C.2 Reactor Top – Characterization Plots

FIGURES C.1 THROUGH C.16		
C.1	REACTOR TOP	TOTAL PLATE COUNT - AEROBIC
C.2	REACTOR TOP	TOTAL PLATE COUNT - ANAEROBIC
C.3	REACTOR TOP	HYDROGEN-SULFIDE PRODUCERS
C.4	REACTOR TOP	CARBOHYDRATE-UTILIZERS - ACID PRODUCERS
C.5	REACTOR TOP	CARBOHYDRATE-UTILIZERS - GAS PRODUCERS
C.6	REACTOR TOP	DENITRIFIERS
C.7	REACTOR TOP	SULFATE REDUCERS
C.8	REACTOR TOP	AMMONIA-NITROGEN
C.9	REACTOR TOP	NITRATE-NITROGEN
C.10	REACTOR TOP	SULFATE
C.11	REACTOR TOP	SULFITE
C.12	REACTOR TOP	pH
C.13	REACTOR TOP	DISSOLVED OXYGEN
C.14	REACTOR TOP	BOD
C.15	REACTOR TOP	TOTAL SUSPENDED SOLIDS
C.16	REACTOR TOP	VOLATILE SUSPENDED SOLIDS

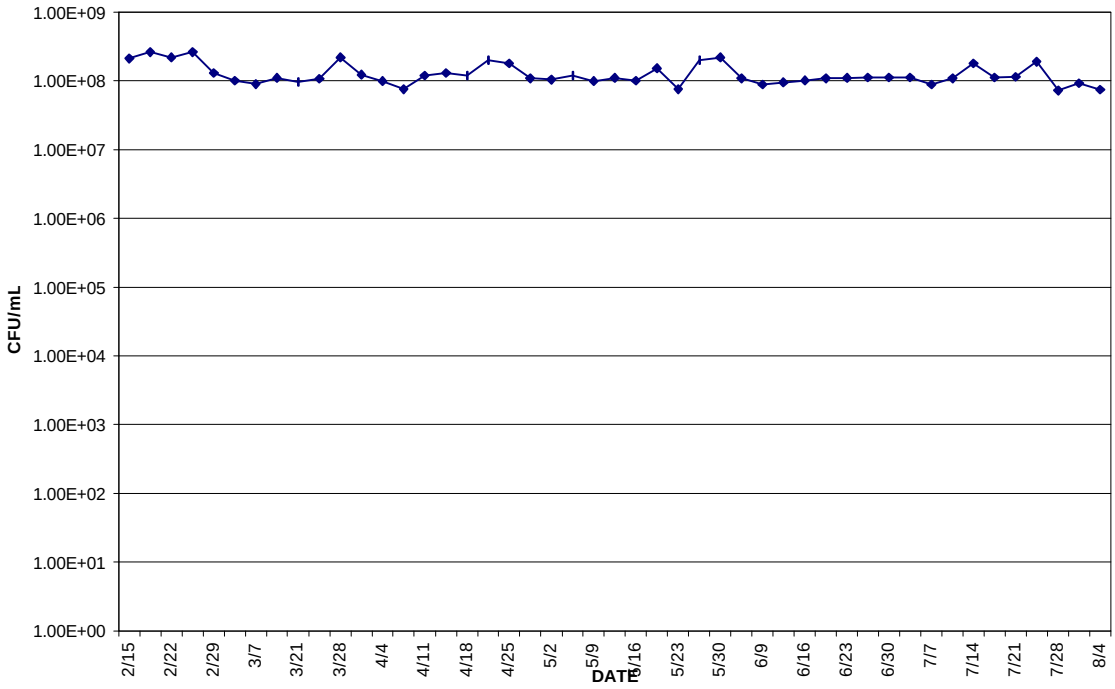

Figure C.1 Reactor top – total plate count - aerobic

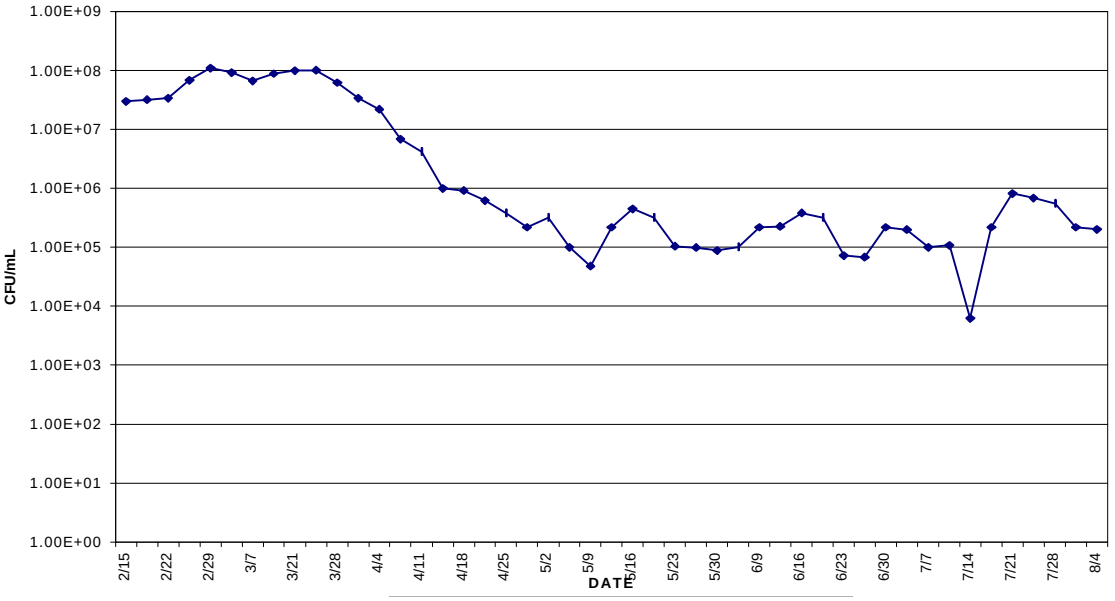

Figure C.2 Reactor top – total plate count - anaerobic

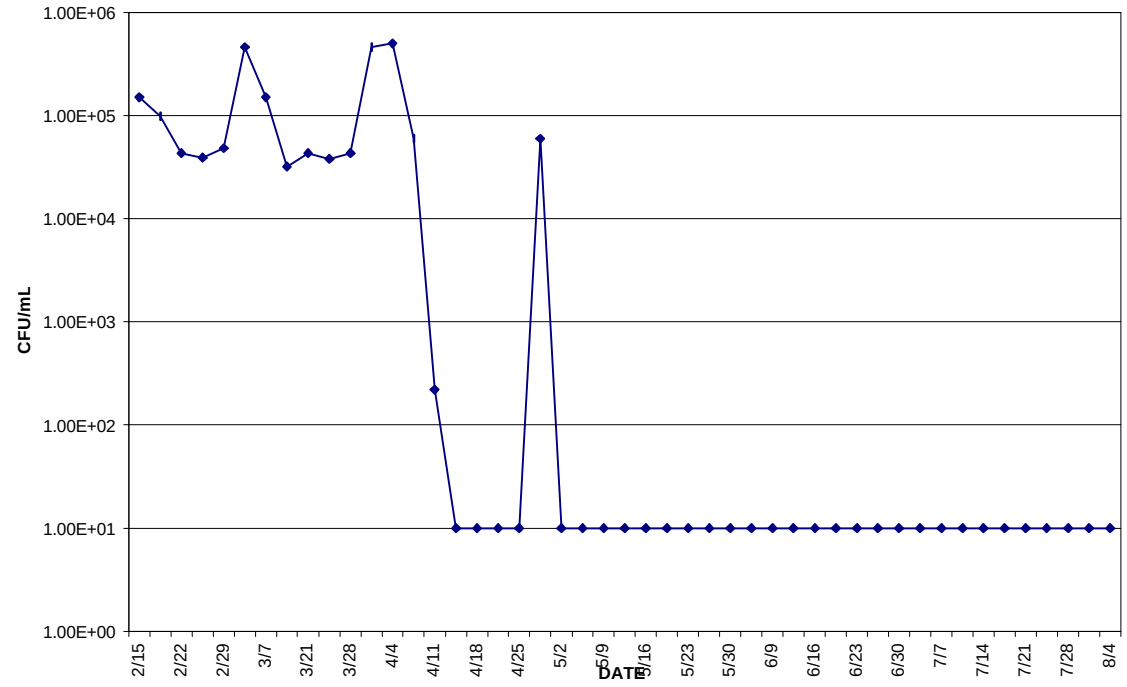

Figure C.3 Reactor top – hydrogen sulfide producers

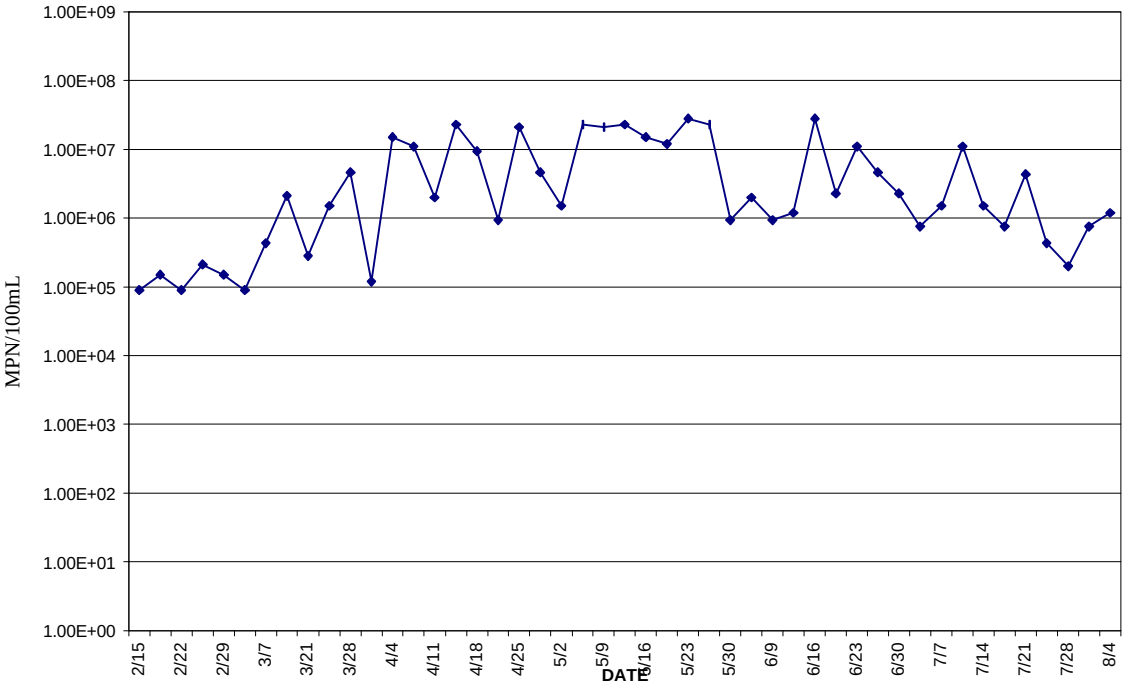

Figure C.4 Reactor top – carbohydrate-utilizers – acid producers

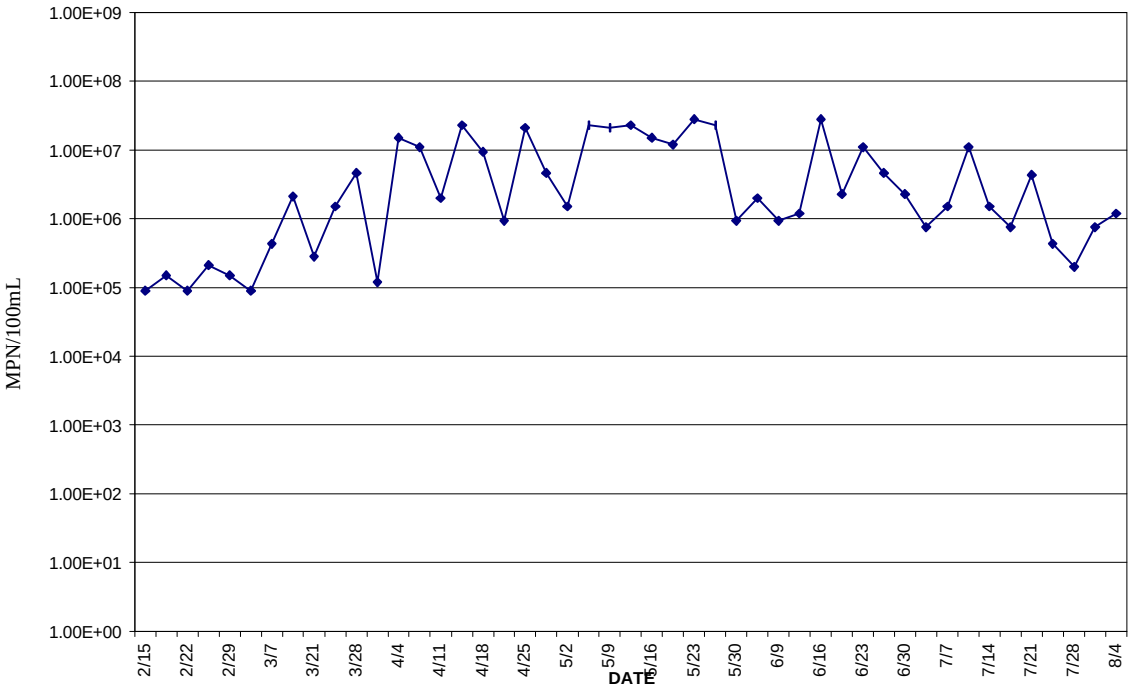

Figure C.5 Reactor top – carbohydrate-utilizers – gas producers

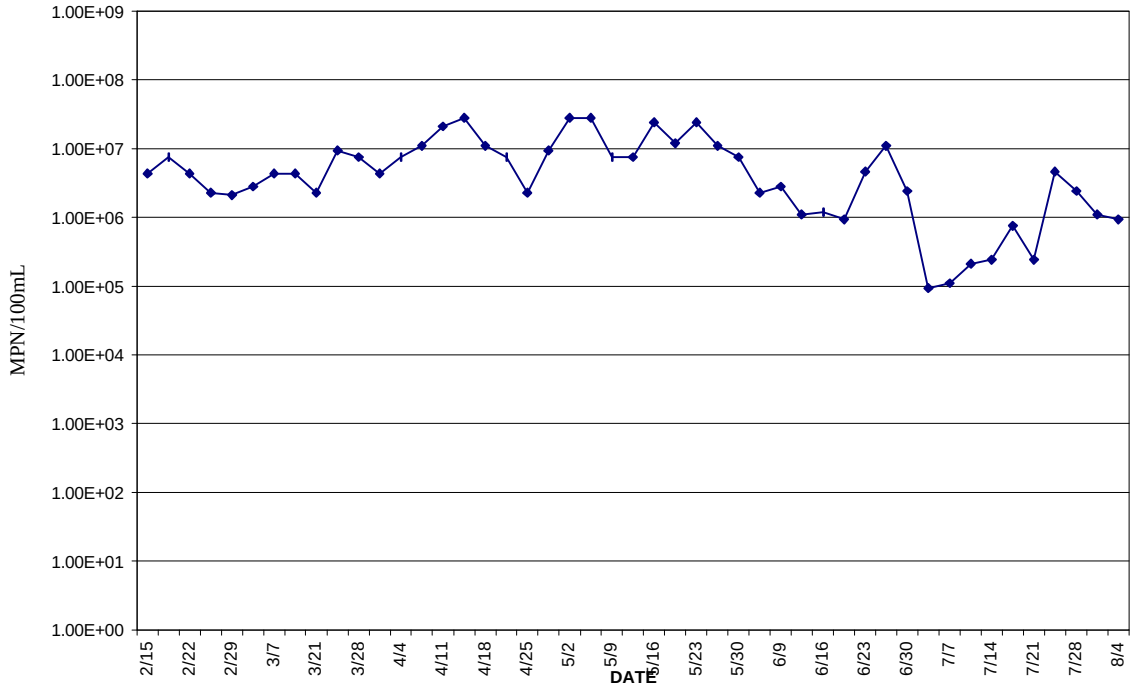

Figure C.6 Reactor top - denitrifiers

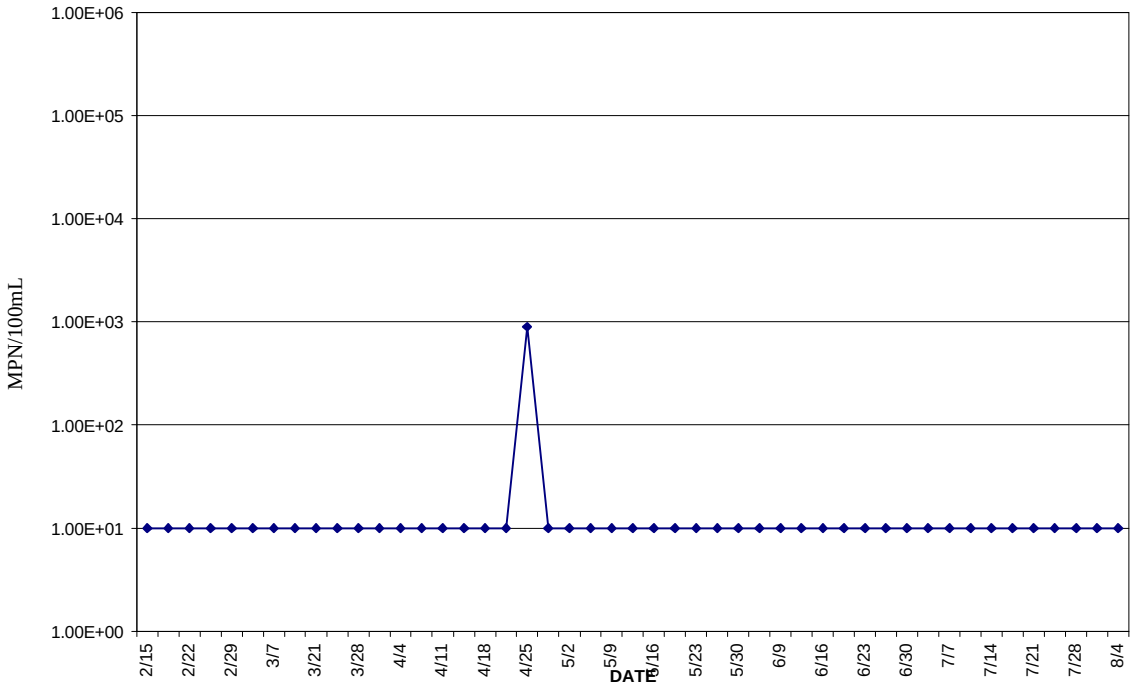

Figure C.7 Reactor top – sulfate reducers

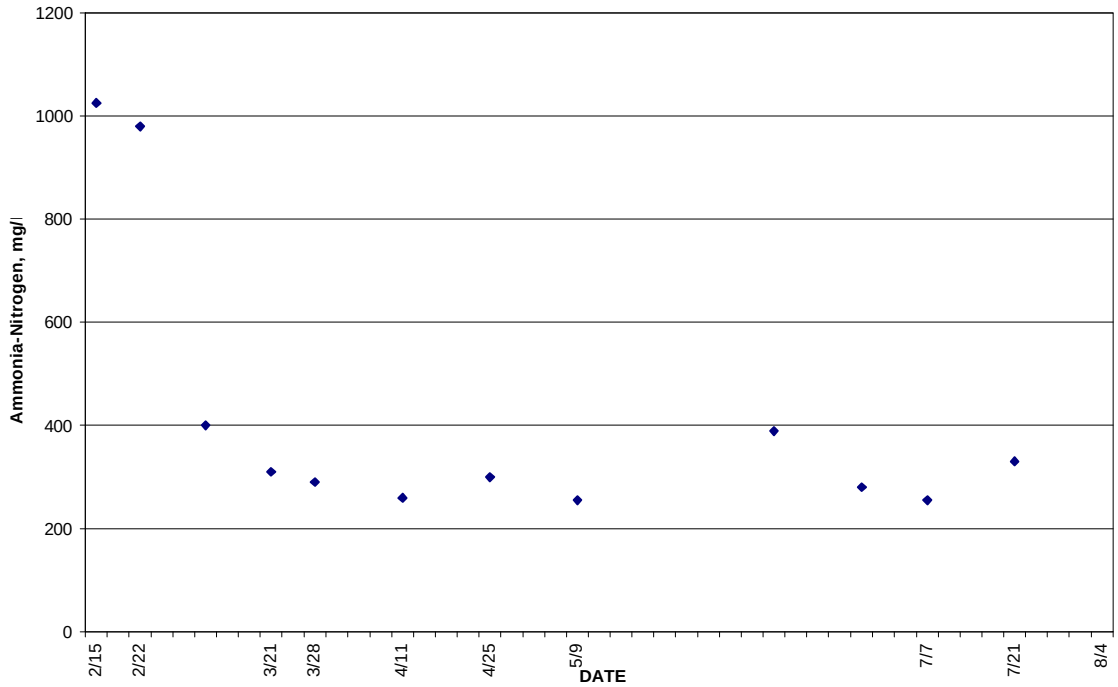

Figure C.8 Reactor top – ammonia-nitrogen

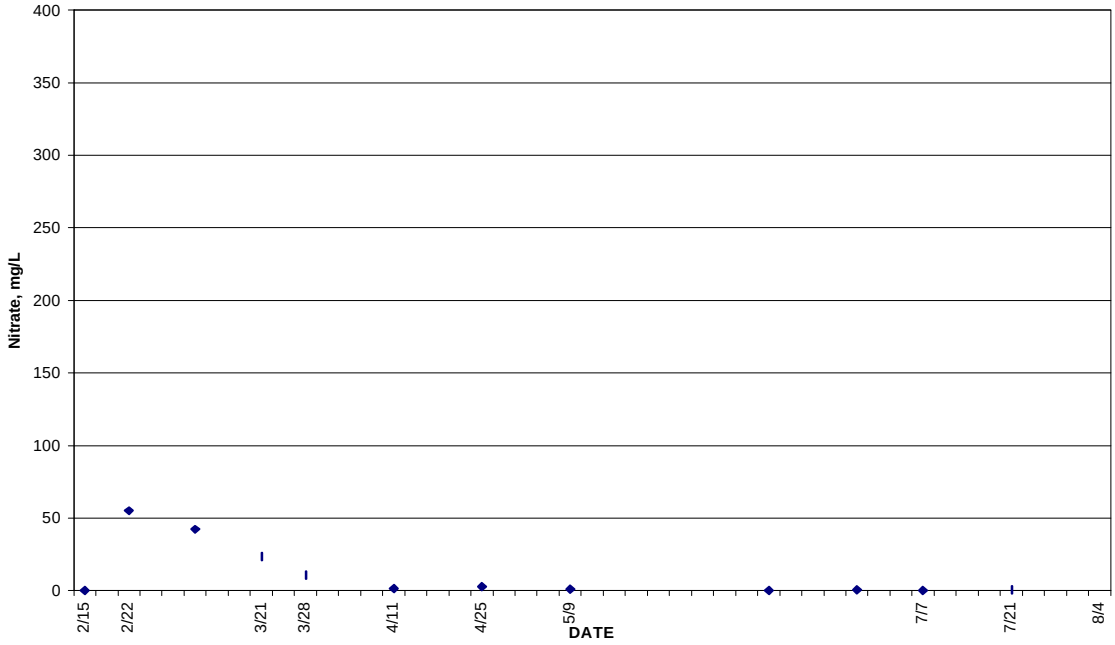

Figure C.9 Reactor top – nitrate-nitrogen

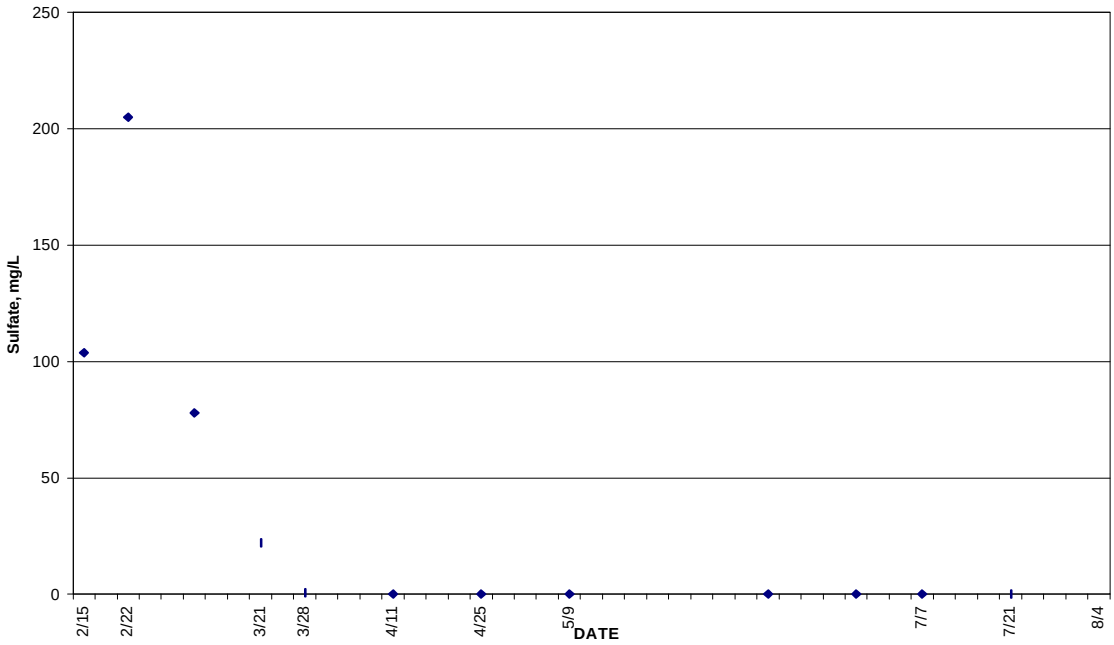

Figure C.10 Reactor top - sulfate

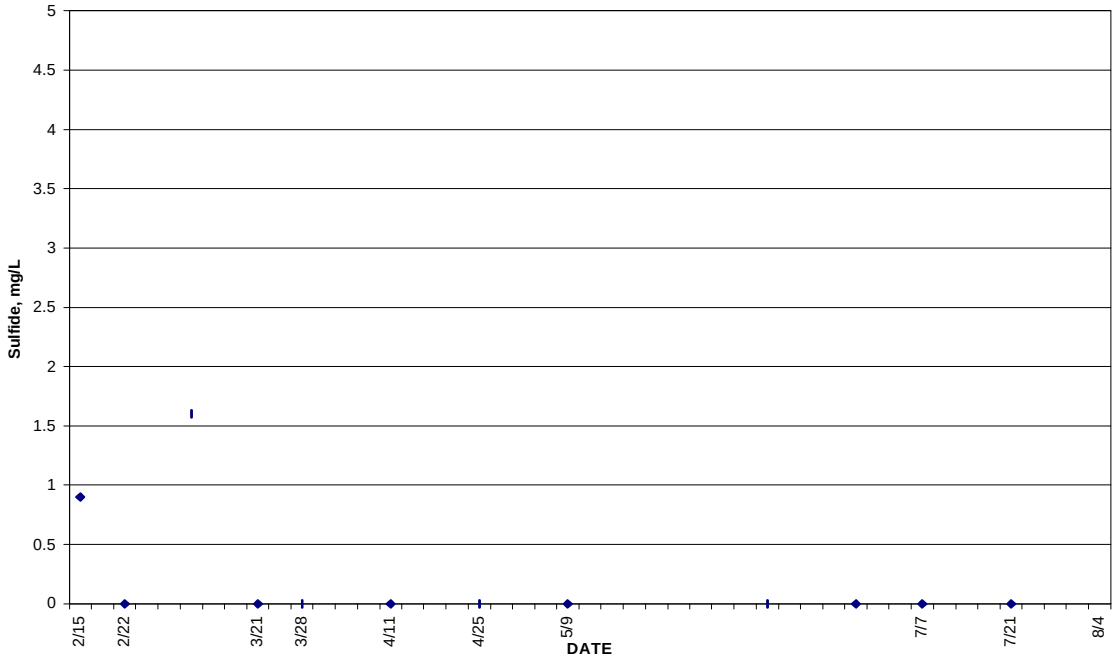

Figure C.11 Reactor top - sulfide

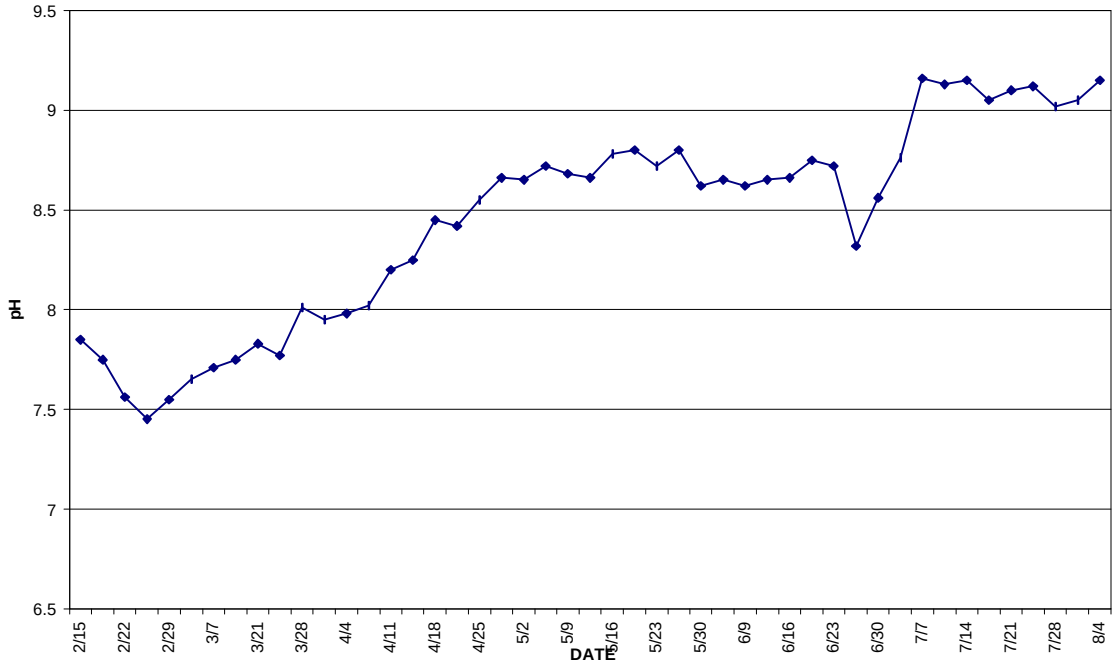

Figure C.12 Reactor top - pH

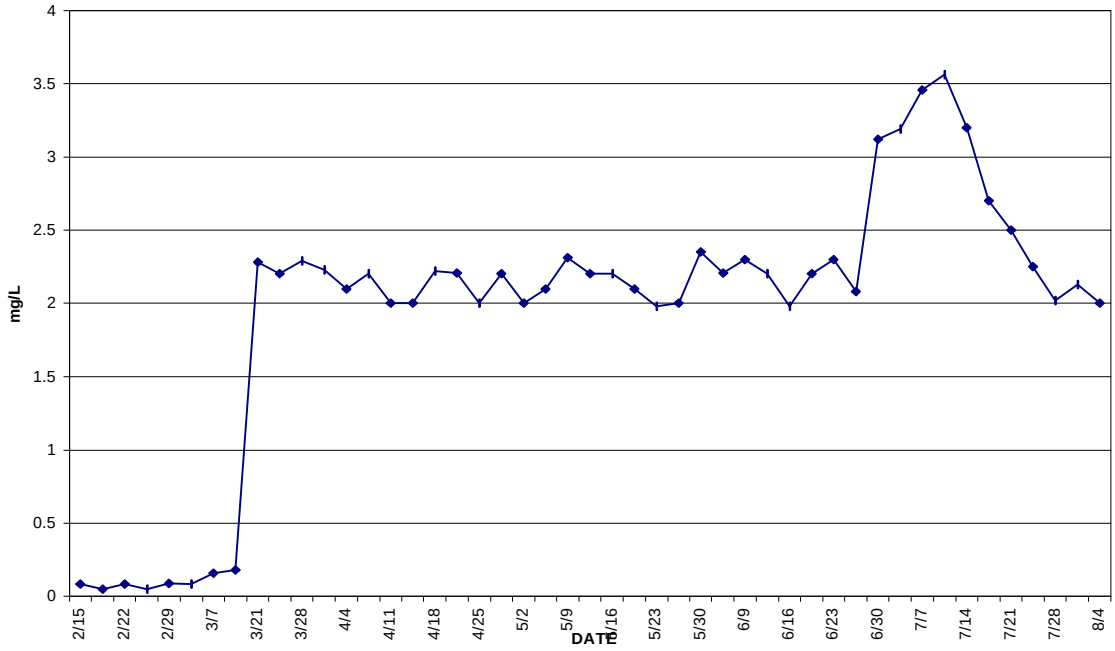

Figure C.13 Reactor top – dissolved oxygen

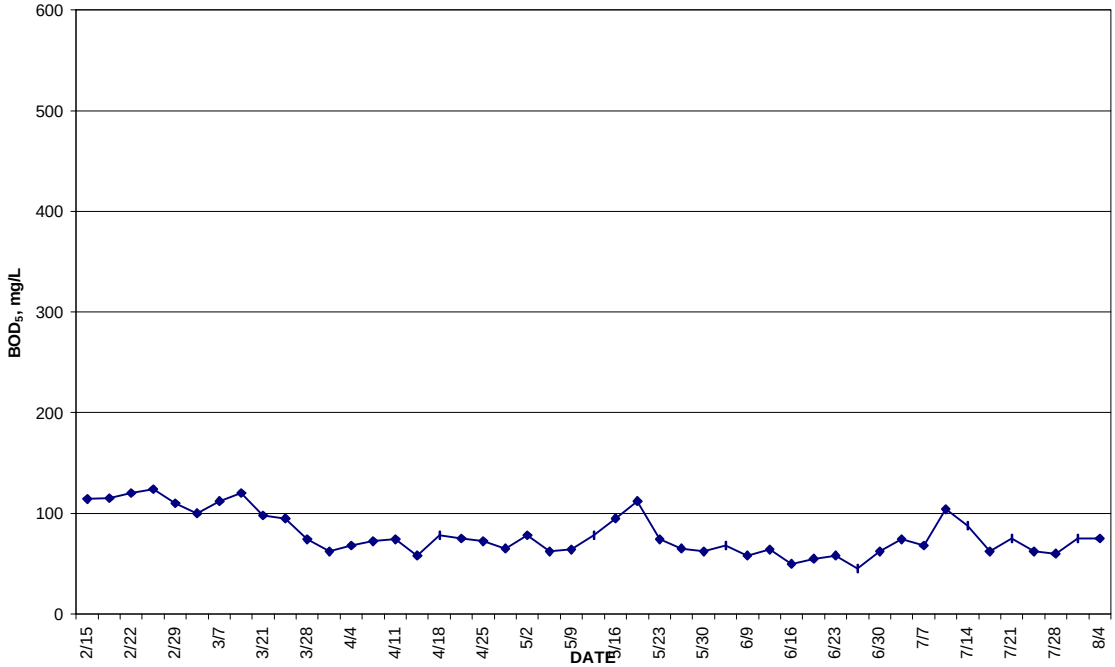

Figure C.14 Reactor top - BOD

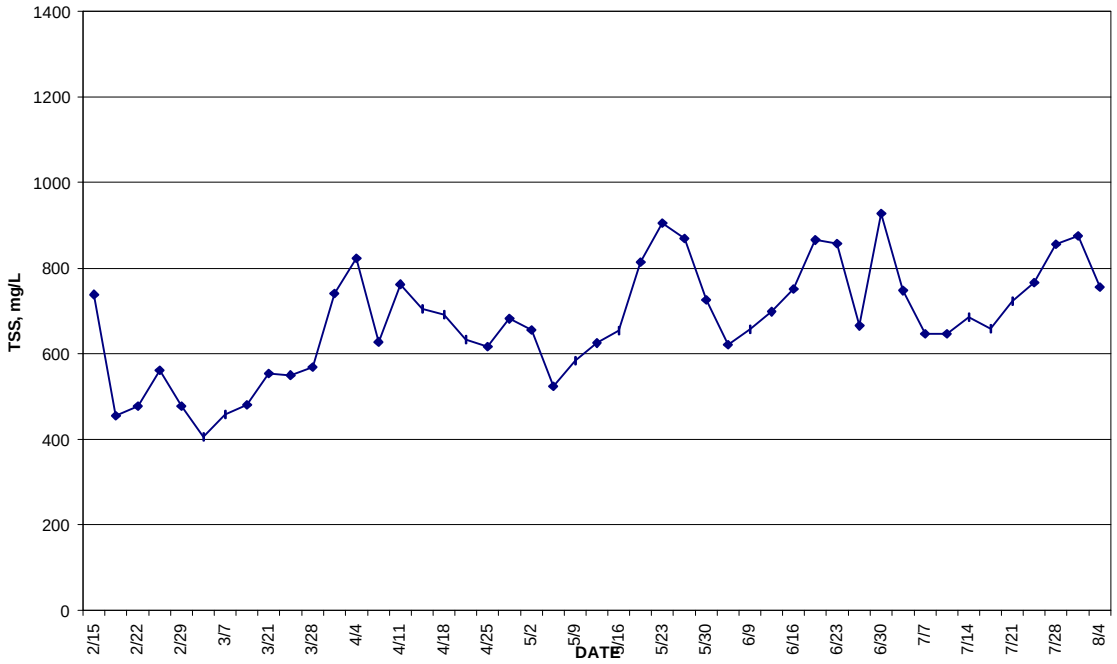

Figure C.15 Reactor top – total suspended solids

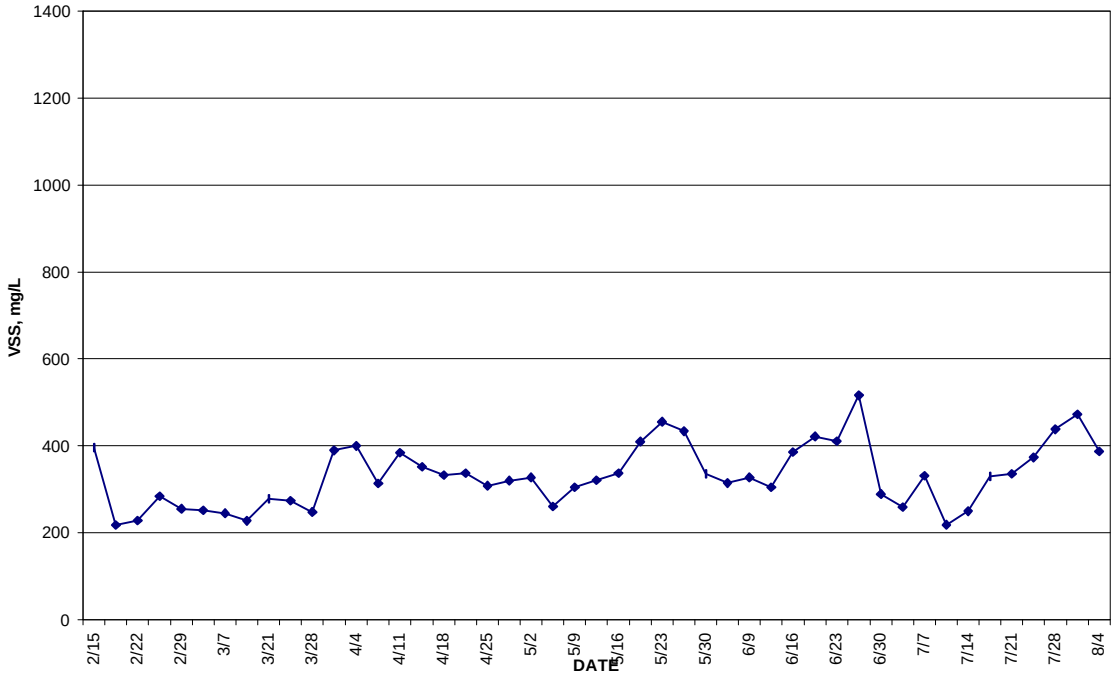

Figure C.16 Reactor top – volatile suspended solids

APPENDIX D

REACTOR TOP vs REACTOR BOTTOM DATA ANALYSIS

Table D.1 Reactor Top vs Reactor Bottom Data Analysis

PARAMETER	2/16	2/18	2/22	2/25	2/29	3/3	3/7	3/17	3/21	3/24	3/28	3/31	4/4	4/7	4/11	4/14	4/18	4/21	4/25	4/28	5/2	5/8	5/9
REACTOR TOP-TOTAL PLATE COUNT-AEROBIC	2.10E+08	2.60E+08	2.20E+08	2.80E+08	1.30E+08	1.00E+08	9.00E+07	1.10E+08	9.60E+07	1.07E+08	2.20E+08	1.22E+08	9.80E+07	7.50E+07	1.20E+08	1.30E+08	1.20E+08	2.00E+08	1.60E+08	1.09E+08	1.04E+08	1.20E+08	9.80E+07
REACTOR BOTTOM-TOTAL PLATE COUNT-AEROBIC	2.00E+08	2.60E+08	2.20E+08	2.80E+08	1.20E+08	1.00E+08	5.00E+07	1.04E+08	4.80E+07	7.00E+07	2.20E+08	1.20E+08	5.60E+07	5.00E+07	1.10E+08	1.30E+08	1.10E+08	2.00E+08	1.60E+08	1.04E+08	9.60E+07	1.20E+08	6.60E+07
REACTOR TOP-TOTAL PLATE COUNT-ANAEROBIC	3.00E+07	3.20E+07	3.40E+07	6.80E+07	1.10E+08	9.20E+07	6.70E+07	8.80E+07	1.00E+08	1.02E+08	6.20E+07	3.40E+07	2.20E+07	6.80E+06	4.20E+06	1.00E+06	9.20E+05	6.20E+05	3.80E+06	2.20E+05	3.20E+05	1.50E+05	4.80E+04
REACTOR BOTTOM-TOTAL PLATE COUNT-ANAEROBIC	3.00E+08	1.50E+08	9.80E+07	7.80E+07	3.30E+08	1.00E+08	7.80E+06	9.20E+07	8.00E+07	5.60E+07	9.80E+07	4.00E+07	4.00E+07	3.20E+07	7.80E+07	5.60E+07	4.00E+07	3.20E+07	4.60E+07	6.20E+07	4.30E+07	7.50E+07	4.40E+07
REACTOR TOP-HYDROGEN SULFIDE PRODUCERS	1.50E+05	9.80E+04	4.30E+04	3.60E+04	4.80E+04	4.60E+05	1.50E+05	3.20E+04	4.30E+04	3.80E+04	4.30E+04	4.60E+05	5.00E+05	6.00E+04	2.20E+02	1.00E+01	1.00E+01	1.00E+01	1.00E+01	6.00E+04	1.00E+01	1.00E+01	1.00E+01
REACTOR BOTTOM-HYDROGEN SULFIDE PRODUCERS	1.50E+05	9.80E+04	4.30E+04	3.60E+04	4.80E+04	4.60E+05	1.50E+05	3.20E+04	4.30E+04	3.80E+04	4.30E+04	4.60E+05	5.00E+05	4.00E+05	4.80E+05	5.00E+05	4.30E+05	6.20E+05	4.00E+04	3.70E+04	1.10E+05	6.00E+05	6.20E+05
REACTOR TOP-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.30E+05	2.10E+06	2.60E+05	1.50E+06	4.60E+06	1.20E+05	1.50E+07	1.10E+07	2.00E+06	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+06	4.30E+05	7.50E+05	9.30E+05	1.20E+06	9.30E+05	1.50E+05	2.10E+06	2.00E+05	2.30E+05	1.10E+06	2.40E+05	1.10E+07	4.60E+06	1.10E+06	1.10E+07	2.40E+07	2.30E+06	1.50E+06	2.40E+06	1.10E+06	1.10E+07	4.60E+07
REACTOR TOP-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.20E+05	2.10E+06	2.60E+05	1.50E+06	4.60E+06	1.20E+05	1.50E+07	1.10E+07	2.00E+06	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	1.10E+06	4.30E+05	7.50E+05	9.30E+05	1.20E+06	9.30E+05	1.50E+05	2.10E+06	2.00E+05	2.30E+05	1.10E+06	9.30E+04	1.10E+07	4.60E+06	1.10E+06	1.10E+07	2.40E+07	2.30E+06	1.50E+06	2.40E+06	1.10E+06	1.10E+07	4.60E+07
REACTOR TOP-DENITRIFIERS	4.30E+06	7.50E+06	4.30E+06	2.30E+06	2.10E+06	2.60E+06	4.30E+06	4.30E+06	2.30E+06	9.30E+06	7.50E+06	4.30E+06	7.50E+06	1.10E+07	2.10E+07	2.80E+07	1.10E+07	7.50E+06	2.30E+06	9.30E+06	2.80E+07	2.80E+07	7.50E+06
REACTOR BOTTOM-DENITRIFIERS	2.30E+06	2.40E+06	4.60E+06	1.10E+07	9.30E+06	1.20E+07	1.10E+07	2.30E+06	2.30E+06	4.60E+06	2.10E+07	1.60E+07	4.30E+07	1.50E+06	2.30E+07	1.50E+06	7.50E+06	2.40E+06	1.10E+06	2.30E+06	2.40E+06	1.50E+06	2.00E+06
REACTOR TOP-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	9.00E+02	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR BOTTOM-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	2.30E+01	4.30E+01	7.50E+01	7.50E+01	9.30E+01	7.50E+01	9.30E+01	2.40E+02	9.30E+02	1.50E+03	9.30E+02	2.10E+03	2.40E+03	1.20E+03
REACTOR TOP-AMMONIA-N	1025		980			400			310		290				260				300				265
REACTOR BOTTOM-AMMONIA-N	1025		980			400			310		290				275				330				310
REACTOR TOP-NITRATE	0.0		55.0			42.0			23.2		10.8				1.2				2.5				0.9
REACTOR BOTTOM-NITRATE	0.0		55.0			42.0			23.2		10.8				8.3				15.5				4.3
REACTOR TOP-SULFATE	104.0		205.0			78.0			22.0		0.6				0.0				0.0				0.0
REACTOR BOTTOM-SULFATE	104.0		205.0			78.0			22.0		18.0				10.0				22.0				14.9
REACTOR TOP-SULFIDE	0.9		0.0			1.8			0.0		0.0				0.0				0.0				0.0
REACTOR BOTTOM-SULFIDE	0.9		0.0			1.8			0.0		0.0				0.0				0.0				0.0
REACTOR TOP-pH	7.85	7.75	7.56	7.45	7.55	7.85	7.71	7.75	7.83	7.77	8.01	7.95	7.98	8.02	8.2	8.25	8.45	8.43	8.55	8.56	8.55	8.72	8.56
REACTOR BOTTOM-pH	7.85	7.75	7.56	7.45	7.55	7.85	7.71	7.75	7.83	7.77	8.01	7.95	7.98	8.15	8.25	8.15	8.35	8.44	8.52	8.72	8.52	8.65	8.56
REACTOR TOP-DISSOLVED OXYGEN	0.09	0.05	0.08	0.05	0.09	0.08	0.16	0.18	2.26	2.2	2.29	2.23	2.1	2.2	2	2	2.22	2.21	2	2.2	2	2.1	2.21
REACTOR BOTTOM-DISSOLVED OXYGEN	0.09	0.05	0.08	0.05	0.09	0.08	0.16	0.18	0.07	0.09	0.11	0.07	0.04	0.08	0.09	0.02	0.19	0.21	0.32	0.16	0.24	0.30	0.31
REACTOR TOP-TEMPERATURE	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	27.2	26.6	26.6	27.1	26.9	26.6	27.1	27.4	27.5	27.4	27.2	26.9
REACTOR BOTTOM-TEMPERATURE	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	26.6	27.2	26.6	26.6	27.1	26.9	26.6	27.1	27.4	27.5	27.4	27.2	26.9
REACTOR TOP-BOD	114	115	120	124	110	100	112	120	96	66	74	62	68	72	74	68	76	75	72	66	76	62	64
REACTOR BOTTOM-BOD	114	115	120	124	110	100	112	120	96	107	95	72	88	105	112	70	98	88	110	88	100	112	70
REACTOR TOP-TSS	730	456	478	562	478	405	456	480	504	550	568	741	823	626	762	705	691	633	617	662	655	523	564
REACTOR BOTTOM-TSS	455	465	520	570	502	423	474	492	564	523	556	754	821	653	569	621	657	519	538	661	522	598	627
REACTOR TOP-VSS	396	218	229	265	255	252	245	228	278	274	348	390	400	314	384	352	333	338	308	320	328	261	305
REACTOR BOTTOM-VSS	255	260	290	325	244	222	265	279	302	260	325	456	450	465	323	340	348	278	305	306	287	323	333

Table D.1 (Con't) Reactor Top vs Reactor Bottom – Data Analysis

PARAMETER	5/12	5/16	5/19	5/23	5/26	5/30	6/6	6/9	6/13	6/16	6/20	6/23	6/27	6/30	7/4	7/7	7/11	7/14	7/18	7/21	7/25	7/28	8/1	8/4
REACTOR TOP-TOTAL PLATE COUNT-AEROBIC	1.10E+08	1.00E+08	1.53E+08	7.50E+07	2.00E+08	2.20E+08	1.08E+08	8.80E+07	9.80E+07	1.02E+08	1.08E+08	1.10E+08	1.12E+08	1.12E+08	1.12E+08	8.60E+07	1.08E+08	1.80E+08	1.12E+08	1.14E+08	1.90E+08	7.20E+07	9.20E+07	7.40E+07
REACTOR BOTTOM-TOTAL PLATE COUNT-AEROBIC	5.00E+07	9.20E+07	1.50E+08	5.50E+07	1.20E+08	1.80E+08	1.04E+08	5.50E+07	5.00E+07	9.20E+07	1.00E+08	9.80E+07	1.04E+08	1.04E+08	9.80E+07	5.50E+07	1.00E+08	1.80E+08	9.80E+07	1.12E+08	1.60E+08	9.80E+07	9.80E+07	5.50E+07
REACTOR TOP-TOTAL PLATE COUNT-ANAEROBIC	2.20E+05	4.60E+06	3.20E+05	1.04E+05	9.60E+04	8.80E+04	1.02E+05	2.20E+06	2.25E+05	3.60E+05	3.20E+05	7.20E+04	6.80E+04	2.20E+05	2.00E+05	1.00E+05	1.08E+05	6.20E+03	2.20E+05	6.20E+05	6.80E+05	5.60E+05	2.20E+05	2.02E+05
REACTOR BOTTOM-TOTAL PLATE COUNT-ANAEROBIC	4.60E+07	4.60E+07	9.80E+07	1.04E+08	3.20E+07	5.50E+07	9.00E+07	4.50E+07	4.60E+07	5.50E+07	9.00E+07	4.50E+07	4.60E+07	4.60E+07	5.50E+07	9.00E+07	1.80E+08	1.20E+08	5.50E+07	9.00E+07	3.50E+07	5.20E+07	6.80E+07	1.04E+07
REACTOR TOP-HYDROGEN SULFIDE PRODUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR BOTTOM-HYDROGEN SULFIDE PRODUCERS	5.50E+04	3.20E+04	2.20E+04	4.60E+05	4.40E+05	6.80E+05	5.00E+05	4.00E+05	9.00E+04	4.60E+05	5.00E+05	3.20E+05	6.80E+04	2.20E+04	2.20E+04	6.80E+04	4.60E+04	2.50E+04	4.60E+04	3.20E+04	2.60E+04	3.00E+04	2.20E+04	3.20E+04
REACTOR TOP-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.60E+07	2.30E+07	9.30E+05	2.00E+06	9.30E+06	1.20E+06	2.60E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.50E+05	1.50E+06	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+05	7.50E+05	1.20E+06
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+06	1.20E+07	9.30E+07	9.30E+07	2.10E+07	9.30E+05	1.20E+06	4.30E+06	9.30E+05	1.10E+07	1.10E+06	2.40E+05	2.10E+06	2.30E+05	4.30E+05	1.10E+06	4.60E+06	2.40E+06	4.30E+05	4.60E+05	2.10E+06	4.30E+05	9.30E+05	9.30E+05
REACTOR TOP-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.60E+07	2.30E+07	9.30E+05	2.00E+06	9.30E+06	1.20E+06	2.60E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.50E+05	1.50E+06	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+05	7.50E+05	1.20E+06
REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	4.60E+07	1.20E+07	9.30E+07	9.30E+07	2.10E+07	9.30E+05	1.20E+06	4.30E+06	9.30E+05	1.10E+07	4.60E+06	2.40E+06	2.10E+06	2.30E+06	4.30E+05	1.10E+06	4.60E+06	2.40E+06	4.30E+05	4.60E+06	2.10E+06	4.30E+05	9.30E+05	9.30E+05
REACTOR TOP-DENITRIFIERS	7.50E+06	2.40E+07	1.20E+07	2.40E+07	1.10E+07	7.50E+06	2.30E+06	2.60E+06	1.10E+06	1.20E+06	9.30E+05	4.60E+06	1.10E+07	2.40E+06	9.30E+04	1.10E+05	2.10E+06	2.40E+05	7.50E+05	2.40E+05	4.60E+06	2.40E+06	1.10E+06	9.30E+05
REACTOR BOTTOM-DENITRIFIERS	1.50E+06	2.30E+06	1.50E+07	9.30E+06	4.60E+06	4.60E+06	2.30E+06	2.30E+06	7.50E+06	4.60E+07	7.50E+06	1.10E+06	2.00E+05	2.10E+05	2.30E+06	1.10E+07	4.60E+07	1.10E+07	4.60E+07	2.30E+06	2.30E+06	9.30E+05	2.40E+05	1.10E+06
REACTOR TOP-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
REACTOR BOTTOM-SULFATE REDUCERS	2.40E+03	4.60E+03	2.40E+03	4.60E+03	1.10E+04	1.10E+04	2.40E+03	9.30E+03	4.60E+03	7.50E+03	9.30E+03	1.10E+04	2.10E+04	2.10E+04	2.00E+04	1.50E+04	9.30E+03	7.50E+03	9.30E+03	7.50E+03	2.40E+02	4.60E+02	1.10E+03	2.10E+02
REACTOR TOP-AMMONIA-N									389				280			255				330				
REACTOR BOTTOM-AMMONIA-N									410				334			310				360				
REACTOR TOP-NITRATE									0.0				0.3			0.0				0.3				
REACTOR BOTTOM-NITRATE									1.8				2.3			0.0				1.1				
REACTOR TOP-SULFATE									0.0				0.0			0.0				0.0				
REACTOR BOTTOM-SULFATE									5.5				14.3			2.3				22.5				
REACTOR TOP-SULFIDE									0.0				0.0			0.0				0.0				
REACTOR BOTTOM-SULFIDE									0.0				0.0			0.0				0.0				
REACTOR TOP-pH	8.66	8.78	8.6	8.72	8.6	8.62	8.66	8.62	8.66	8.66	8.75	8.72	8.32	8.56	8.76	9.16	9.13	9.15	9.05	9.1	9.12	9.02	9.05	9.15
REACTOR BOTTOM-pH	8.44	8.45	8.56	8.65	8.67	8.69	8.56	8.56	8.62	8.65	8.38	8.52	8.25	8.47	8.64	8.99	8.89	8.99	8.64	8.85	8.92	8.89	8.95	8.92
REACTOR TOP-DISSOLVED OXYGEN	2.2	2.2	2.1	1.98	2	2.35	2.21	2.3	2.2	1.98	2.2	2.3	2.08	2.12	3.19	3.46	3.88	3.2	2.7	2.5	2.28	2.02	2.13	2
REACTOR BOTTOM-DISSOLVED OXYGEN	0.25	0.21	0.16	0.14	0.09	0.09	0.13	0.21	0.16	0.18	0.20	0.22	0.43	0.61	0.76	0.29	0.68	0.54	0.32	0.65	0.55	0.45	0.41	0.23
REACTOR TOP-TEMPERATURE	27.5	27.9	27.8	27.6	28	27.6	27.5	28.4	28.5	29.4	29.6	30.3	31.2	30	30	30	30	29.5	29.8	29.4	30	30	30.1	29.8
REACTOR BOTTOM-TEMPERATURE	27.5	27.9	27.8	27.6	28	27.6	27.5	28.4	28.5	29.4	29.6	30.2	31.2	30	30	30	30	29.5	29.8	29.4	30	30	30.1	29.8
REACTOR TOP-BOD	78	95	112	74	66	82	68	58	64	50	55	58	45	62	74	66	104	68	62	75	62	60	75	75
REACTOR BOTTOM-BOD	95	112	120	88	75	88	76	62	76	56	66	61	60	64	109	95	150	132	84	87	78	70	114	88
REACTOR TOP-TSS	625	654	614	605	669	726	621	657	698	751	666	667	665	628	749	647	647	665	659	723	766	696	675	766
REACTOR BOTTOM-TSS	664	673	526	550	466	490	661	662	560	669	648	668	528	664	589	532	531	521	660	632	590	649	560	678
REACTOR TOP-VSS	321	338	410	456	434	336	315	328	305	366	422	411	516	269	259	331	219	250	332	336	373	439	473	387
REACTOR BOTTOM-VSS	365	325	266	286	276	255	303	312	321	387	390	378	285	466	323	302	245	274	366	350	333	380	364	318

Table D.2 Correlation Coefficient – Reactor Top vs Reactor Bottom

PARAMETER	CORRELATION COEFFICIENT
REACTOR TOP-TOTAL PLATE COUNT-AEROBIC **REACTOR BOTTOM-TOTAL PLATE COUNT-AEROBIC**	0.93
REACTOR TOP-TOTAL PLATE COUNT-ANAEROBIC **REACTOR BOTTOM-TOTAL PLATE COUNT-ANAEROBIC**	0.34
REACTOR TOP-HYDROGEN SULFIDE PRODUCERS **REACTOR BOTTOM-HYDROGEN SULFIDE PRODUCERS**	0.23
REACTOR TOP-CARBOHYDRATE UTILIZERS-ACID PRODUCERS **REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-ACID PRODUCERS**	0.60
REACTOR TOP-CARBOHYDRATE UTILIZERS-GAS PRODUCERS **REACTOR BOTTOM-CARBOHYDRATE UTILIZERS-GAS PRODUCERS**	0.59
REACTOR TOP-DENITRIFIERS **REACTOR BOTTOM-DENITRIFIERS**	-0.16
REACTOR TOP-SULFATE REDUCERS **REACTOR BOTTOM-SULFATE REDUCERS**	-0.07
REACTOR TOP-AMMONIA-N **REACTOR BOTTOM-AMMONIA-N**	1.00
REACTOR TOP-NITRATE **REACTOR BOTTOM-NITRATE**	0.98
REACTOR TOP-SULFATE **REACTOR BOTTOM-SULFATE**	0.99
REACTOR TOP-SULFIDE **REACTOR BOTTOM-SULFIDE**	1.00
REACTOR TOP-pH **REACTOR BOTTOM-pH**	0.98
REACTOR TOP-DISSOLVED OXYGEN **REACTOR BOTTOM-DISSOLVED OXYGEN**	0.55
REACTOR TOP-TEMPERATURE **REACTOR BOTTOM-TEMPERATURE**	1.00
REACTOR TOP-BOD **REACTOR BOTTOM-BOD**	0.79
REACTOR TOP-TSS **REACTOR BOTTOM-TSS**	0.41
REACTOR TOP-VSS **REACTOR BOTTOM-VSS**	0.17

Table D.3 Reactor Top vs Bottom – Characterization Plots

FIGURES D.1 THROUGH D.16		
D.1	REACTOR TOP vs BOTTOM	TOTAL PLATE COUNT - AEROBIC
D.2	REACTOR TOP vs BOTTOM	TOTAL PLATE COUNT - ANAEROBIC
D.3	REACTOR TOP vs BOTTOM	HYDROGEN-SULFIDE PRODUCERS
D.4	REACTOR TOP vs BOTTOM	CARBOHYDRATE-UTILIZERS - ACID PRODUCERS
D.5	REACTOR TOP vs BOTTOM	CARBOHYDRATE-UTILIZERS - GAS PRODUCERS
D.6	REACTOR TOP vs BOTTOM	DENITRIFIERS
D.7	REACTOR TOP vs BOTTOM	SULFATE REDUCERS
D.8	REACTOR TOP vs BOTTOM	AMMONIA-NITROGEN
D.9	REACTOR TOP vs BOTTOM	NITRATE-NITROGEN
D.10	REACTOR TOP vs BOTTOM	SULFATE
D.11	REACTOR TOP vs BOTTOM	SULFITE
D.12	REACTOR TOP vs BOTTOM	pH
D.13	REACTOR TOP vs BOTTOM	DISSOLVED OXYGEN
D.14	REACTOR TOP vs BOTTOM	BOD
D.15	REACTOR TOP vs BOTTOM	TOTAL SUSPENDED SOLIDS
D.16	REACTOR TOP vs BOTTOM	VOLATILE SUSPENDED SOLIDS

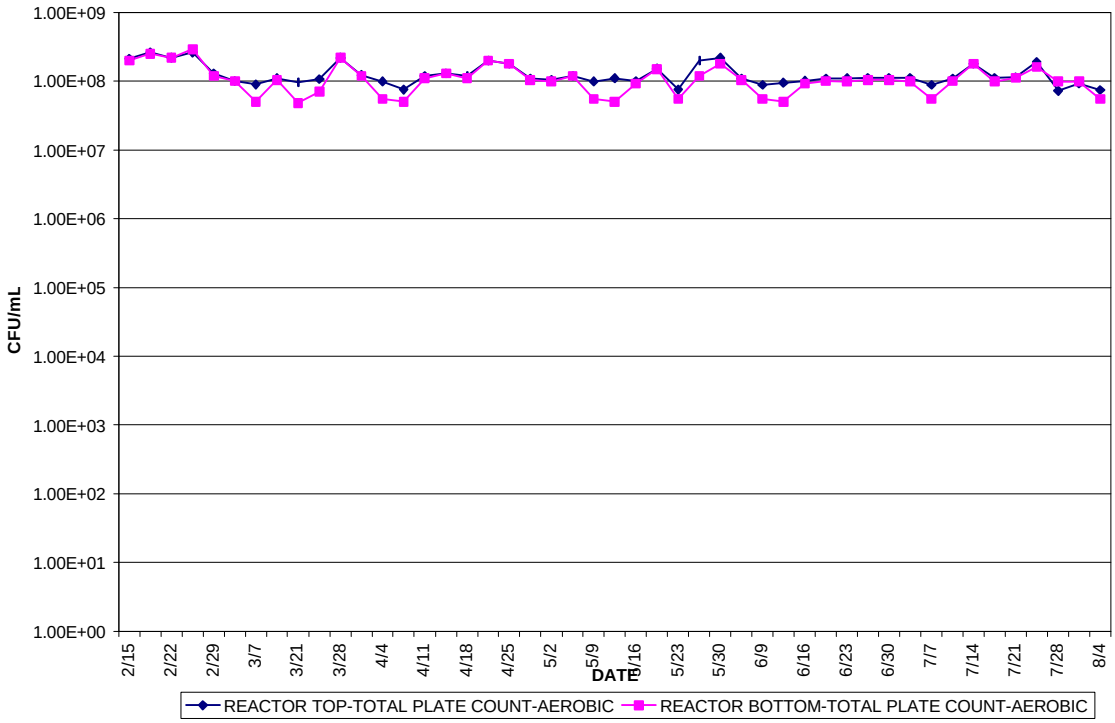

Figure D.1 Reactor top vs bottom – total plate counts - aerobic

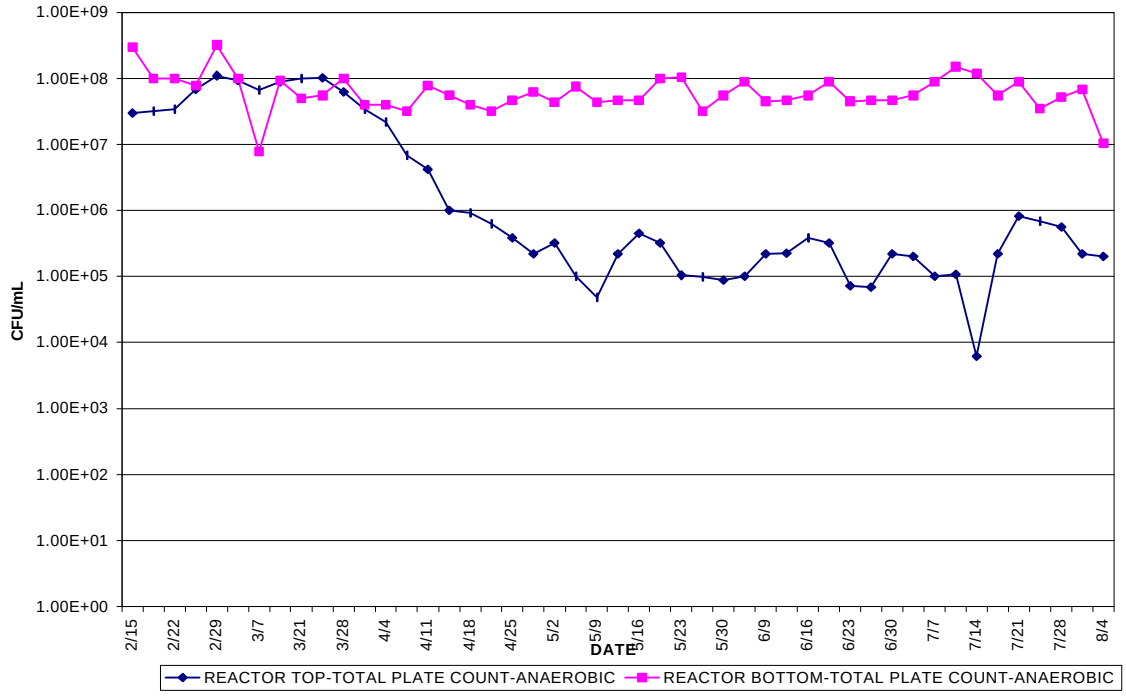

Figure D.2 Reactor top vs bottom – total plate counts - anaerobic

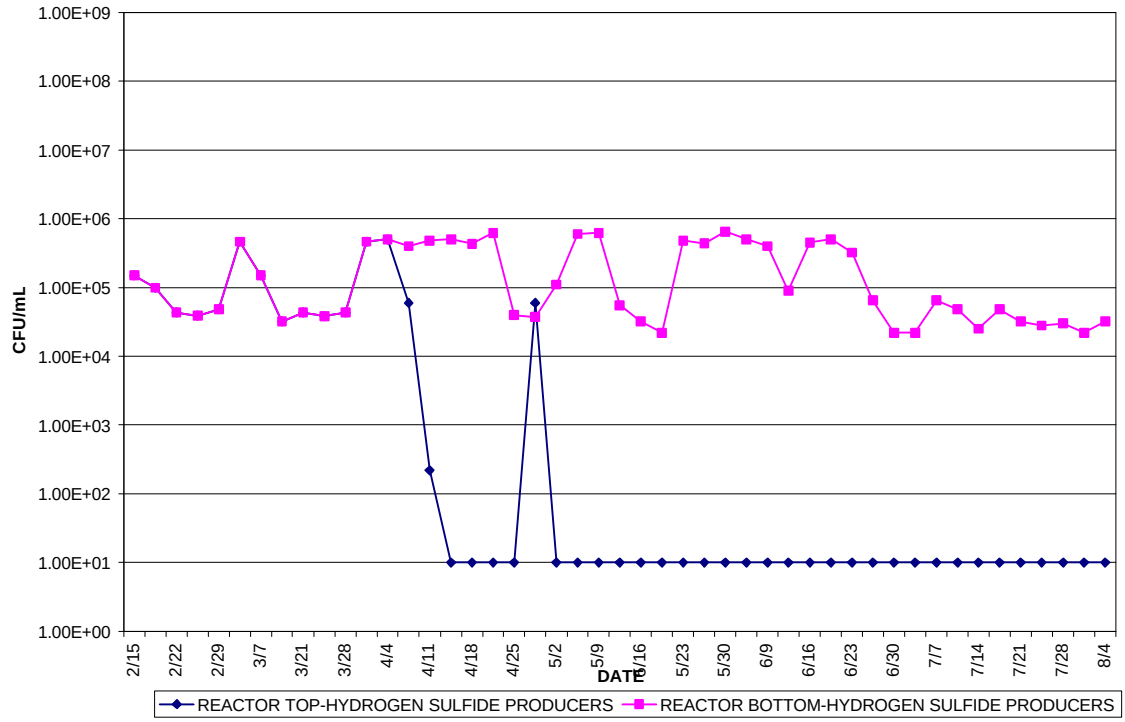

Figure D.3 Reactor top vs bottom – hydrogen-sulfide producers

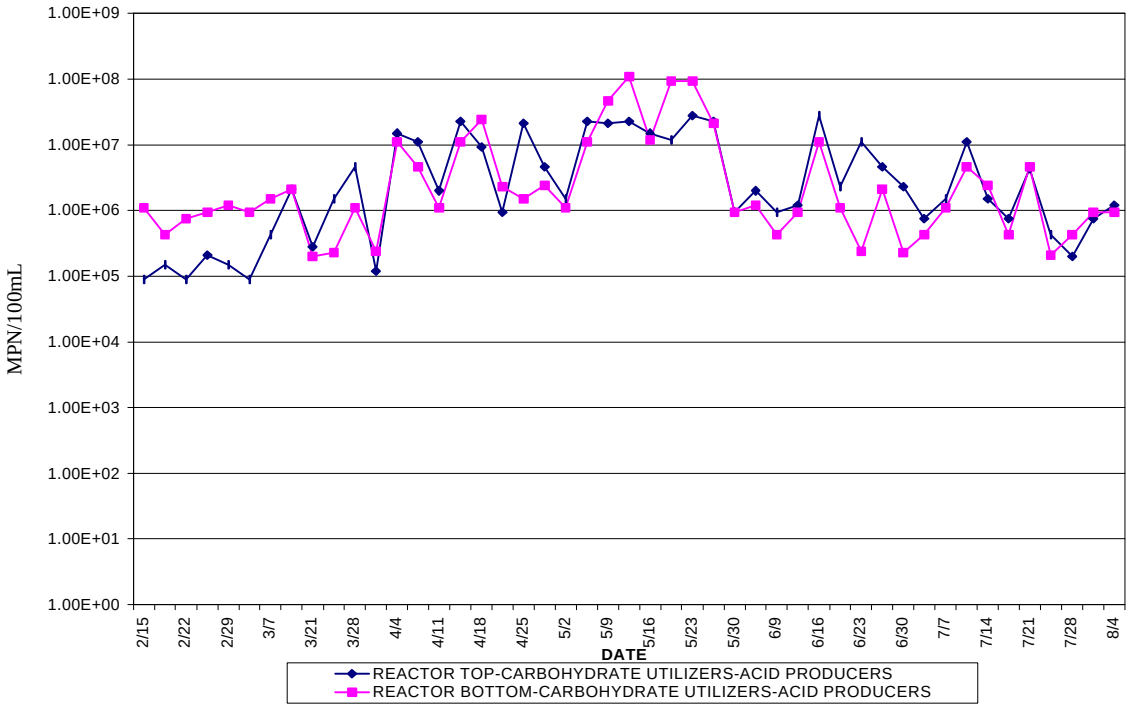

Figure D.4 Reactor top vs bottom – carbohydrate-utilizers – acid-producers

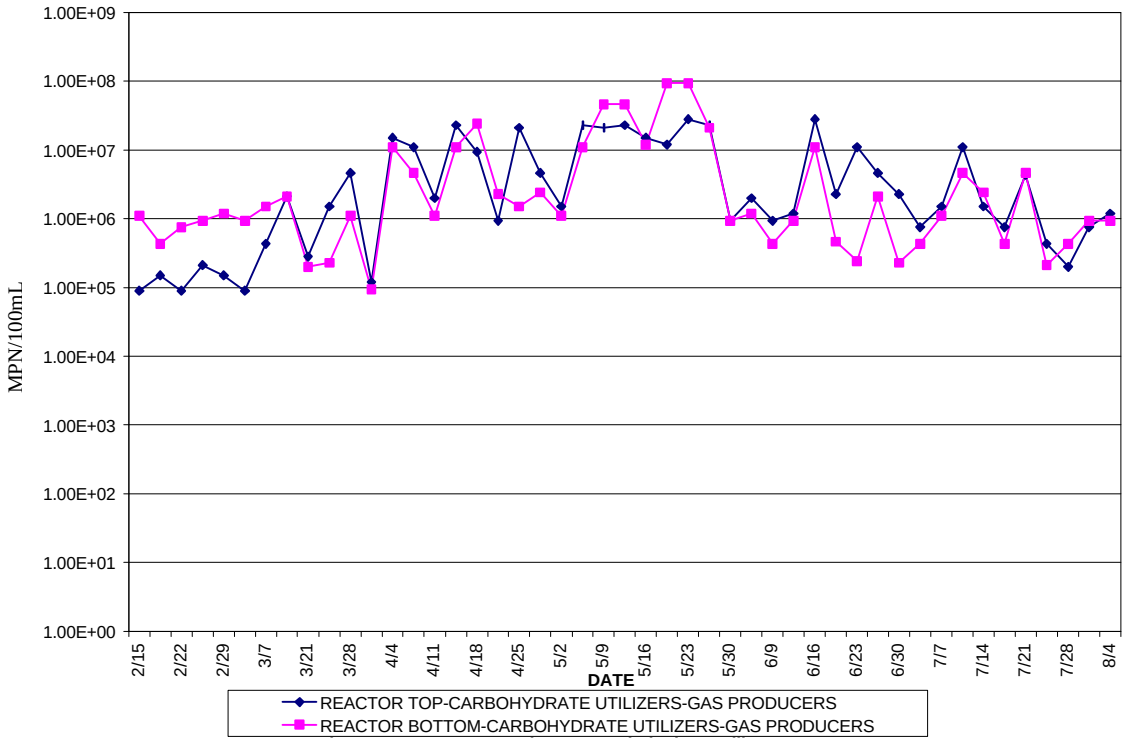

Figure D.5 Reactor top vs bottom – carbohydrate-utilizers – gas producers

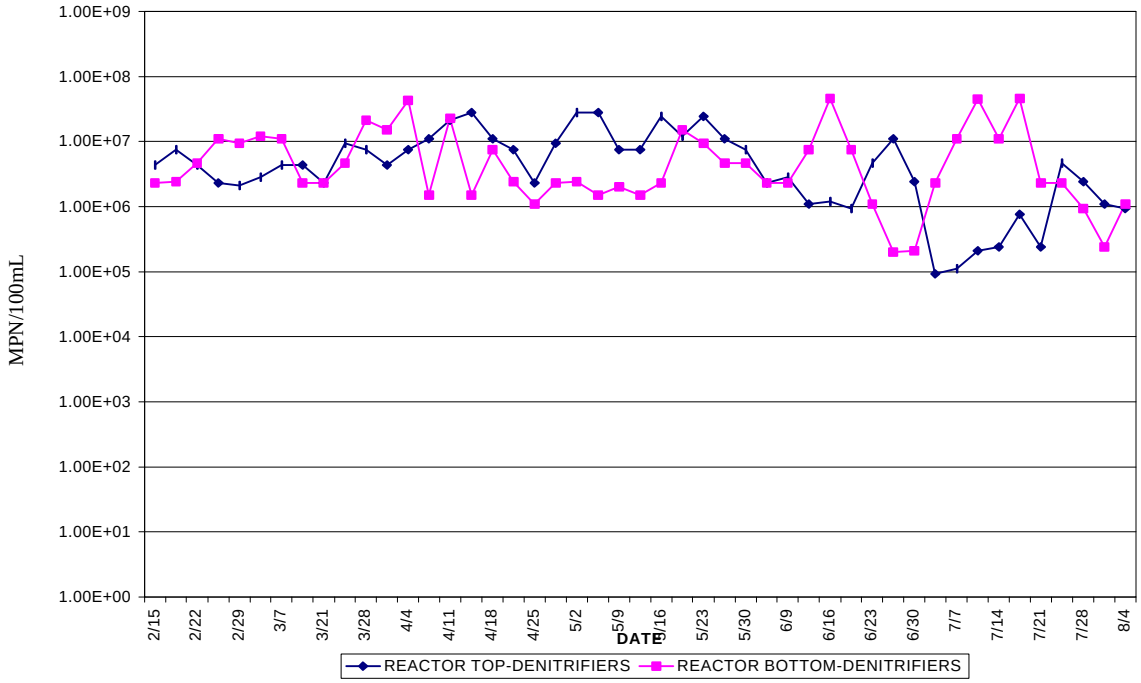

Figure D.6 Reactor top vs bottom - denitrifiers

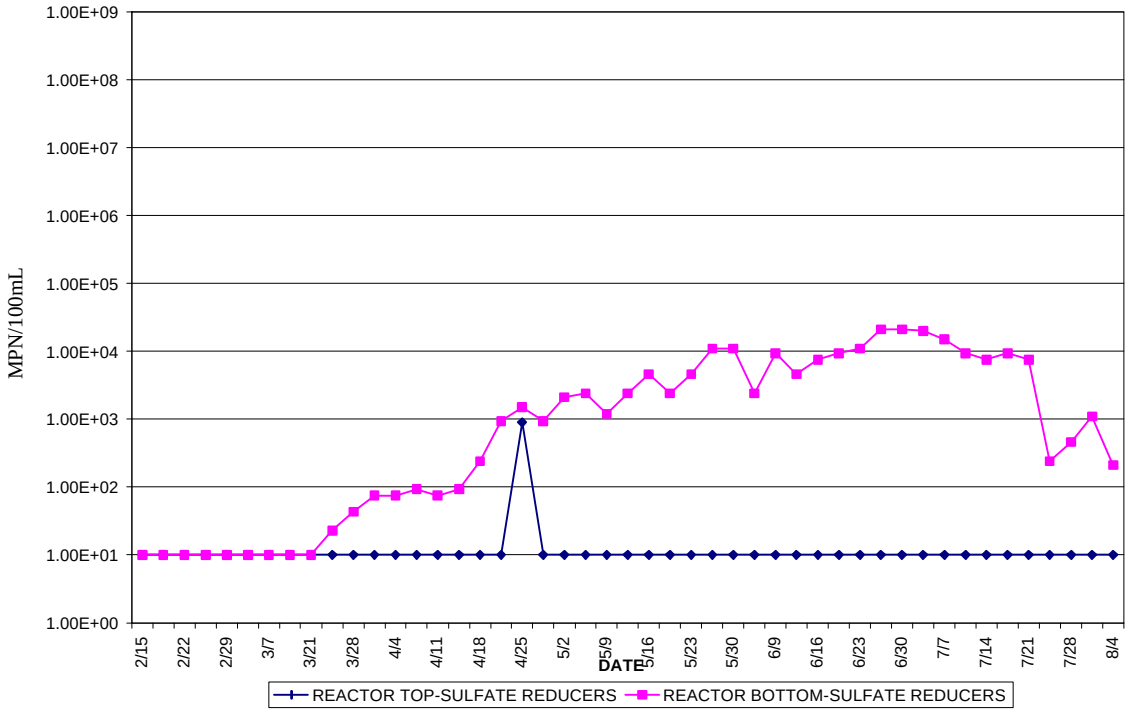

Figure D.7 Reactor top vs bottom – sulfate reducers

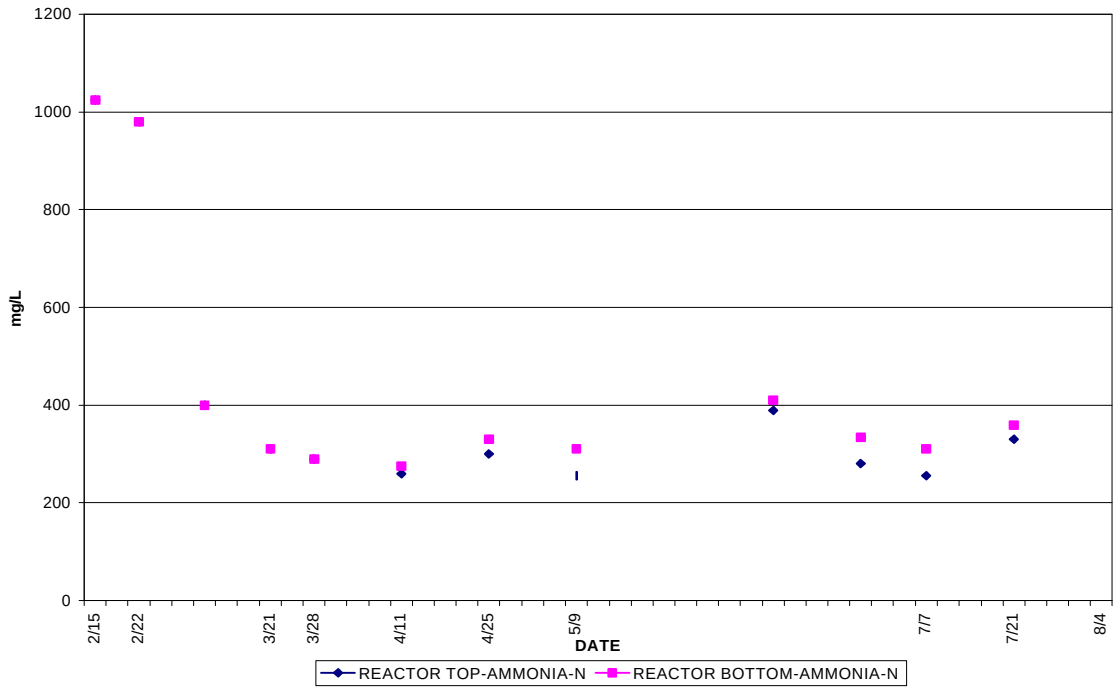

Figure D.8 Reactor top vs bottom – ammonia-nitrogen

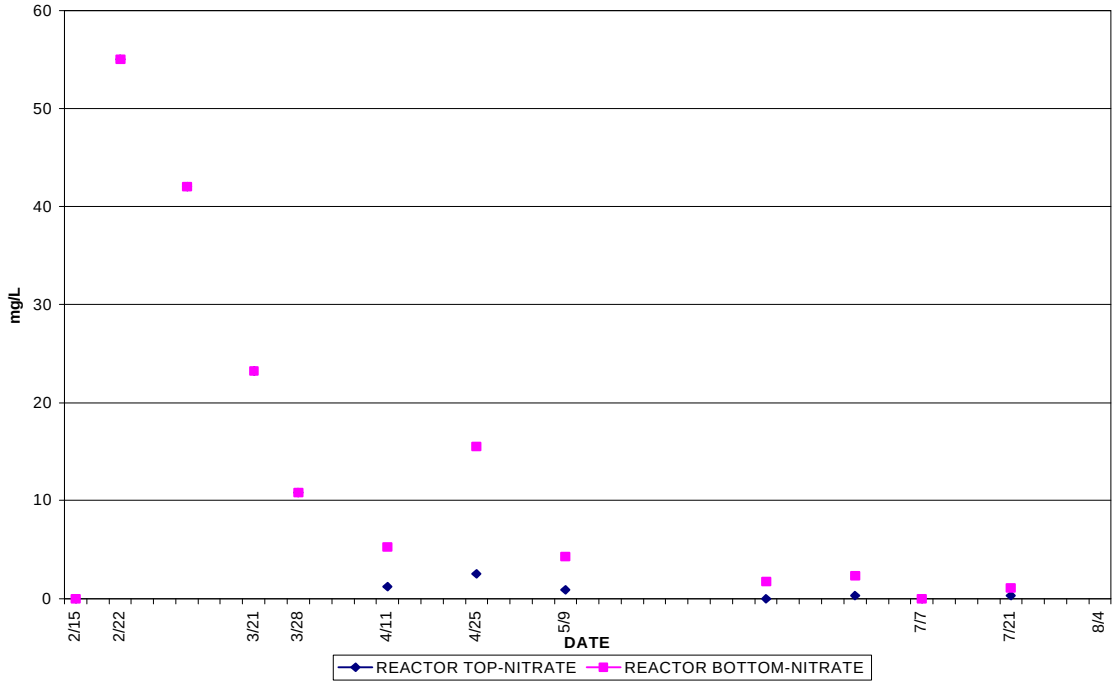

Figure D.9 Reactor top vs bottom – nitrate-nitrogen

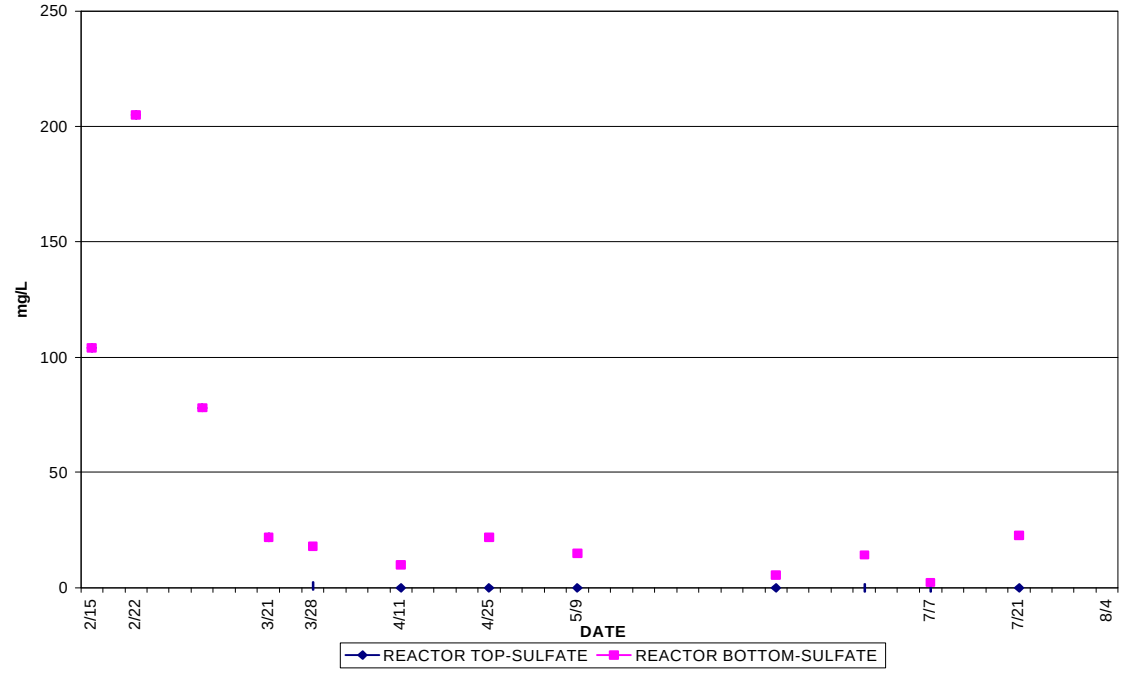

Figure D.10 Reactor top vs bottom - sulfate

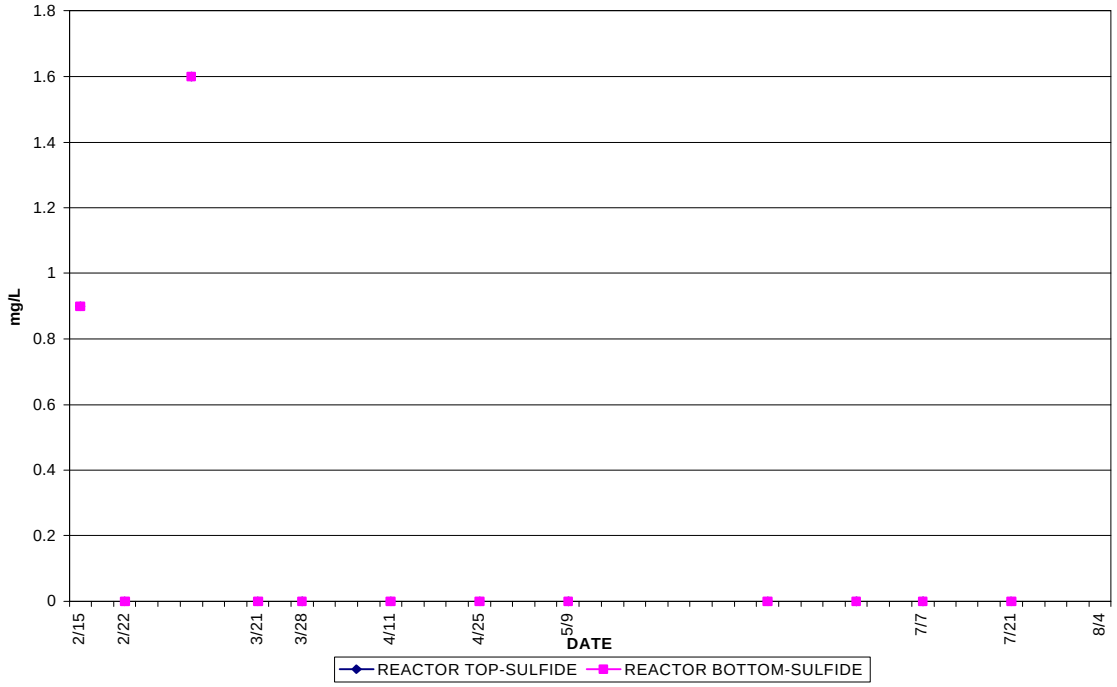

Figure D.11 Reactor top vs bottom - sulfide

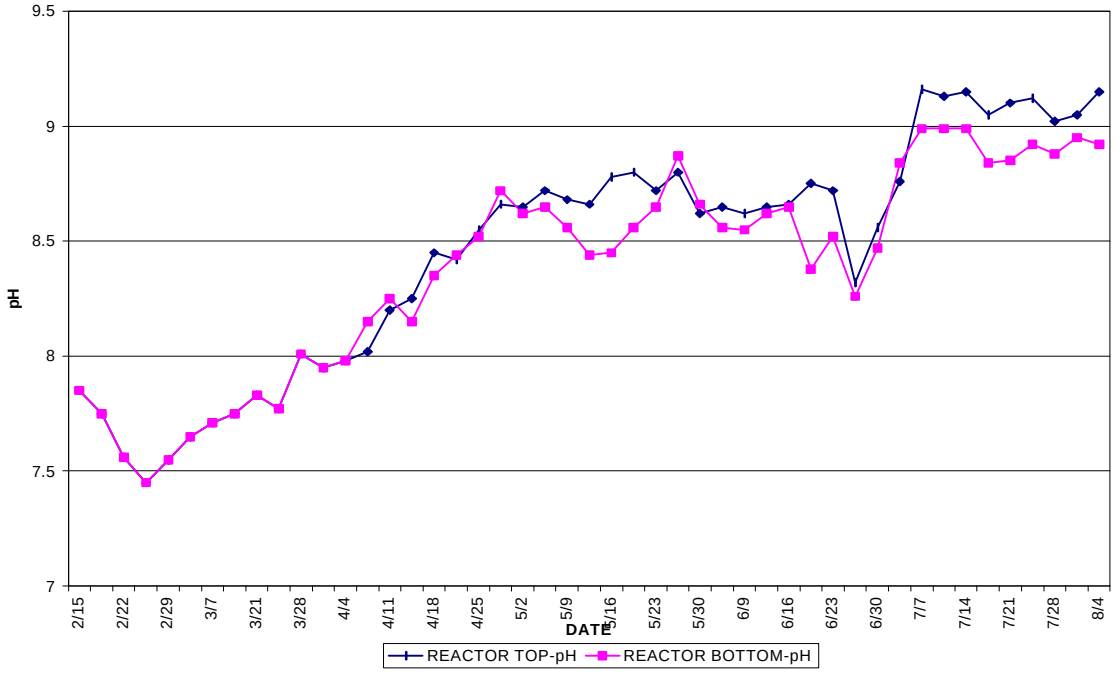

Figure D.12 Reactor top vs bottom - pH

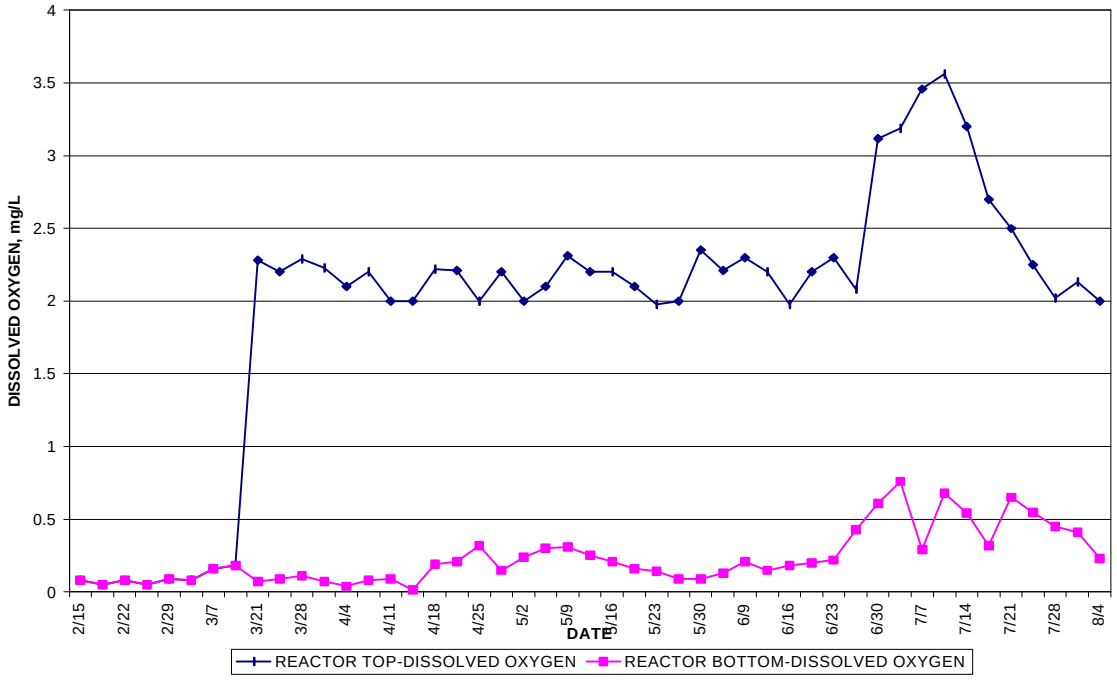

Figure D.13 Reactor top vs bottom – dissolved oxygen

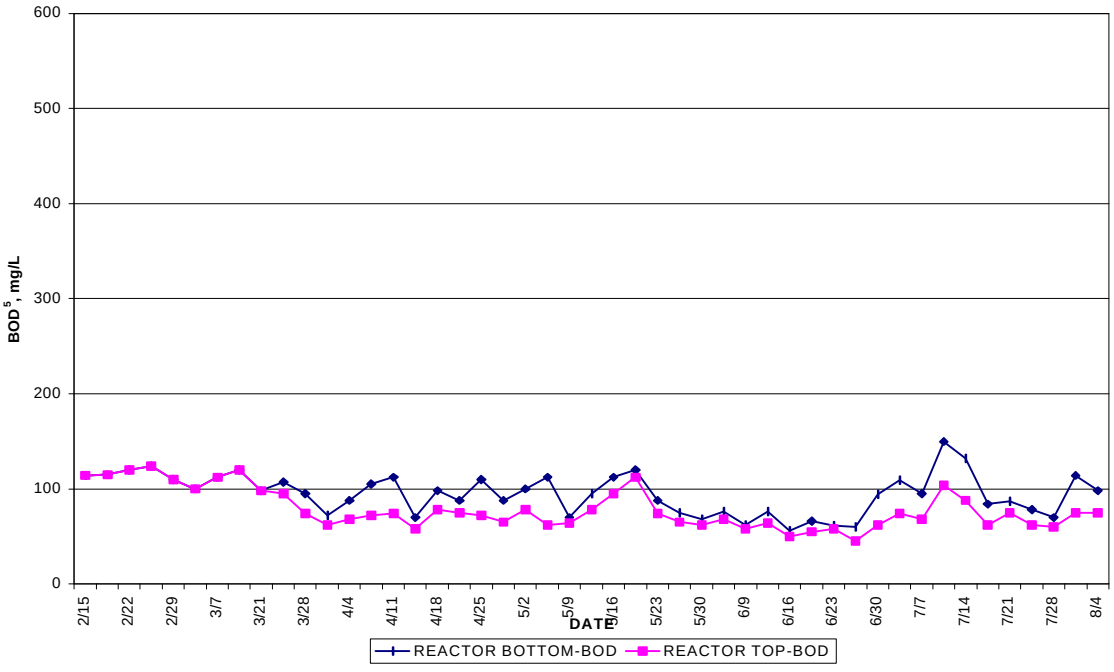

Figure D.14 Reactor top vs bottom - BOD

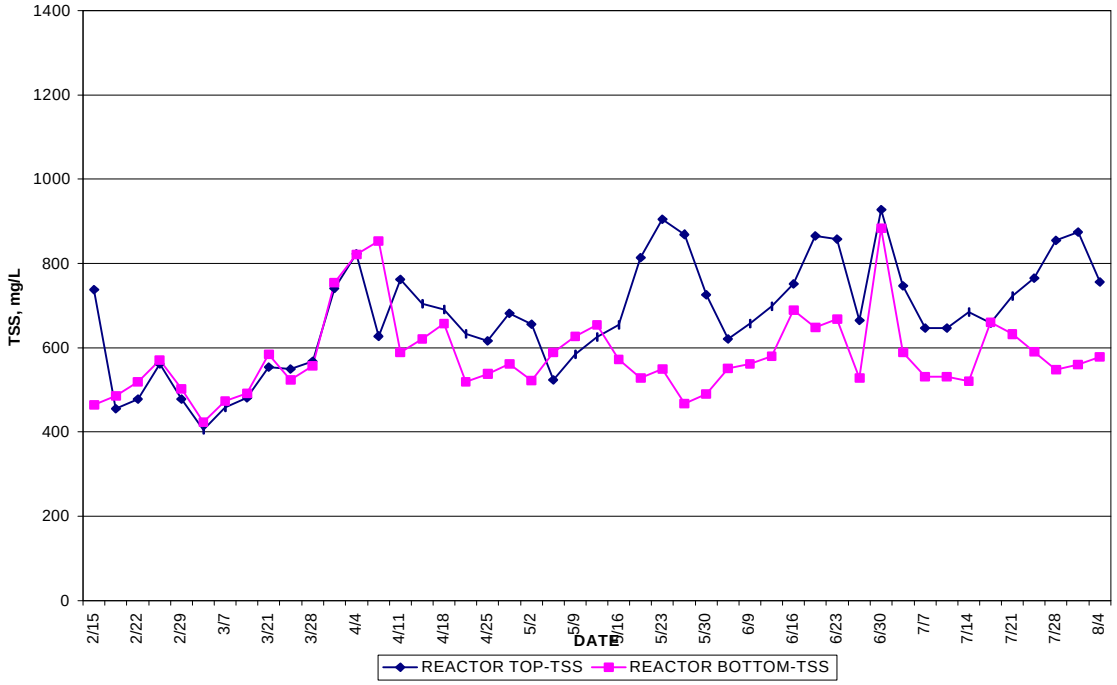

Figure D.15 Reactor top vs bottom – total suspended solids

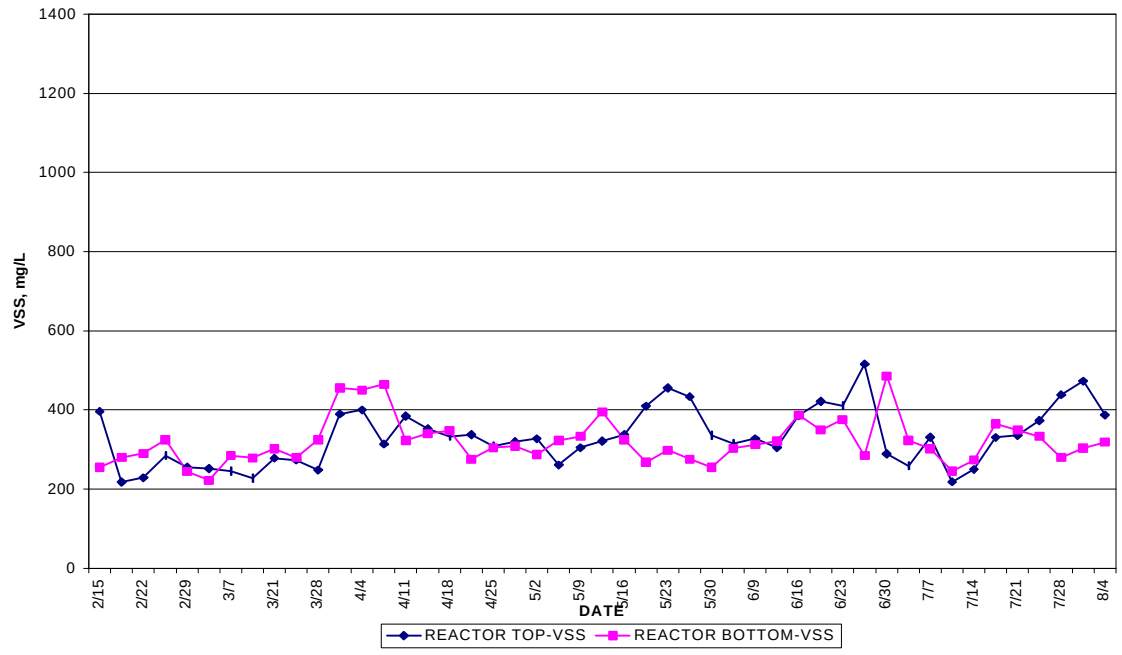

Figure D.16 Reactor top vs bottom – volatile suspended solids

APPENDIX E

INFLUENT vs EFFLUENT DATA ANALYSIS

Table E.1 Influent vs Effluent – Data Analysis

PARAMETER	3/16	3/19	3/22	3/25	3/29	3/5	3/7	3/17	3/21	3/24	3/28	3/31	4/4	4/7	4/11	4/14	4/18	4/21	4/28	4/28	5/2	5/5	5/9
INFLUENT-TOTAL PLATE COUNT-AEROBIC	1.23E+07	5.50E+07	1.10E+07	1.50E+07	4.80E+07	9.50E+07	1.00E+07	1.10E+07	1.10E+07	1.50E+07	1.10E+08	2.30E+08	1.50E+07	1.50E+07	5.00E+07	1.10E+07	4.80E+07	9.30E+07	1.23E+07	2.30E+07	1.00E+07	1.00E+07	2.30E+08
EFFLUENT-TOTAL PLATE COUNT-AEROBIC	2.10E+08	2.60E+08	2.20E+08	2.60E+08	1.30E+08	1.00E+08	9.00E+07	1.10E+08	9.60E+07	1.07E+08	2.20E+08	1.22E+08	9.80E+07	7.50E+07	1.20E+08	1.30E+08	1.20E+08	2.00E+08	1.60E+08	1.06E+08	1.04E+08	1.20E+08	9.80E+07
INFLUENT-TOTAL PLATE COUNT-ANAEROBIC	2.30E+06	3.50E+07	3.10E+06	9.00E+06	9.20E+06	3.70E+07	6.00E+06	6.00E+06	5.80E+06	1.10E+07	3.20E+07	1.10E+08	4.80E+06	9.50E+06	3.90E+07	8.50E+06	2.90E+07	7.80E+07	1.50E+07	1.00E+07	5.80E+06	7.20E+06	1.90E+07
EFFLUENT-TOTAL PLATE COUNT-ANAEROBIC	3.00E+07	3.20E+07	3.40E+07	6.80E+07	1.10E+08	9.20E+07	8.70E+07	8.80E+07	1.00E+08	1.02E+08	6.20E+07	3.40E+07	2.20E+07	6.80E+06	4.20E+06	1.00E+06	9.20E+05	6.20E+05	3.80E+05	2.20E+05	3.20E+05	1.00E+05	4.80E+04
INFLUENT-HYDROGEN SULFIDE PRODUCERS	2.10E+04	3.20E+04	3.50E+04	6.00E+04	5.20E+04	3.00E+04	5.50E+04	4.80E+05	9.00E+05	4.40E+05	4.60E+05	3.20E+06	9.00E+06	5.50E+06	4.60E+06	3.20E+05	8.00E+05	1.00E+06	7.80E+05	3.20E+05	9.00E+04	8.50E+04	4.80E+04
EFFLUENT-HYDROGEN SULFIDE PRODUCERS	1.50E+06	9.80E+04	4.30E+04	3.90E+04	4.80E+04	4.60E+05	1.50E+05	3.20E+04	4.30E+04	3.80E+04	4.30E+04	4.60E+05	5.00E+06	6.00E+04	2.20E+02	1.00E+01	1.00E+01	1.00E+01	1.00E+01	6.00E+04	1.00E+01	1.00E+01	1.00E+01
INFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+07	4.60E+06	1.10E+07	2.10E+06	4.30E+05	9.30E+05	4.30E+05	2.00E+05	7.80E+06	1.10E+07	1.10E+07	4.60E+06	1.10E+07	2.10E+06	2.30E+05	2.10E+05	1.10E+07	1.20E+06	1.10E+07	1.10E+07	1.10E+07	4.60E+06	1.10E+07
EFFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.30E+05	2.10E+06	2.80E+05	1.50E+05	4.60E+06	1.20E+06	1.50E+07	1.10E+07	2.00E+06	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07
INFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	1.10E+07	4.60E+06	1.10E+07	2.10E+06	4.30E+05	9.30E+05	4.30E+05	2.00E+06	7.80E+05	1.10E+07	4.60E+06	4.60E+06	1.10E+07	2.10E+06	2.30E+06	2.10E+05	1.10E+07	1.20E+06	1.10E+07	1.10E+07	1.10E+07	4.60E+06	2.10E+06
EFFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	9.00E+04	1.50E+05	9.00E+04	2.10E+05	1.50E+05	9.00E+04	4.30E+05	2.10E+06	2.80E+05	1.50E+06	4.60E+06	1.20E+06	1.50E+07	1.10E+07	2.00E+06	2.30E+07	9.30E+06	9.30E+05	2.10E+07	4.60E+06	1.50E+06	2.30E+07	2.10E+07
INFLUENT-DENITRIFIERS	2.80E+05	5.40E+05	7.50E+05	1.20E+06	1.10E+07	4.50E+06	1.10E+06	4.80E+06	2.40E+07	2.00E+06	2.30E+06	1.20E+07	1.50E+07	1.50E+06	2.10E+06	1.10E+06	4.50E+07	2.10E+06	4.80E+06	2.10E+07	4.30E+06	4.30E+05	9.30E+06
EFFLUENT-DENITRIFIERS	4.30E+06	7.50E+06	4.30E+06	2.30E+06	2.10E+06	2.80E+06	4.30E+06	4.30E+06	2.30E+06	9.30E+05	7.50E+06	4.30E+06	7.50E+06	1.10E+07	2.10E+07	2.80E+07	1.10E+07	7.50E+06	2.30E+06	9.30E+06	2.80E+07	2.80E+07	7.50E+06
INFLUENT-SULFATE REDUCERS	7.50E+03	1.50E+04	2.10E+04	1.10E+05	2.10E+05	4.60E+05	1.10E+06	2.10E+06	9.30E+05	7.50E+05	1.10E+06	4.60E+05	2.80E+04	4.30E+04	1.50E+04	7.50E+04	9.30E+03	7.50E+03	2.00E+04	7.50E+04	9.30E+04	2.10E+04	2.80E+04
EFFLUENT-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	9.00E+02	1.00E+01	1.00E+01	1.00E+01	1.00E+01
INFLUENT-AMMONIA-N	1025		980			400			355		270				680				880				400
EFFLUENT-AMMONIA-N	1025		980			400			310		290				260				300				285
INFLUENT-NITRATE	0.0		55.0			42.0			56.0		42.0				17.0				160.0				42.0
EFFLUENT-NITRATE	0.0		55.0			42.0			23.2		10.8				1.2				2.5				0.9
INFLUENT-SULFATE	104.0		205.0			78.0			116.0		96.5				178.0				58.0				98.0
EFFLUENT-SULFATE	104.0		205.0			78.0			22.0		0.6				0.0				0.0				0.0
INFLUENT-SULFIDE	0.9		0.0			1.6			0.0		0.0				0.0				0.0				0.6
EFFLUENT-SULFIDE	0.9		0.0			1.6			0.0		0.0				0.0				0.0				0.0
INFLUENT-pH	7.84	7.76	7.21	6.98	6.87	7.14	7.02	7.12	7.24	7.56	7.11	6.99	6.85	7.01	7.14	7.24	7.16	7.54	7.50	7.52	7.14	7.35	7.00
EFFLUENT-pH	7.85	7.76	7.56	7.45	7.55	7.66	7.71	7.76	7.83	7.77	8.01	7.96	7.98	8.02	8.2	8.25	8.45	8.42	8.55	8.66	8.65	8.72	8.58
INFLUENT-DISSOLVED OXYGEN	0.05	0.05	0.04	0.05	0.04	0.05	0.03	0.04	0.02	0.02	0.05	0.05	0.07	0.05	0.03	0.04	0.02	0.02	0.03	0.05	0.04	0.05	0.06
EFFLUENT-DISSOLVED OXYGEN	0.06	0.05	0.06	0.05	0.09	0.06	0.16	0.16	2.26	2.2	2.29	2.23	2.1	2.2	2	2	2.22	2.21	2	2.2	2	2.1	2.31
INFLUENT-TEMPERATURE	24.0	24.2	24.6	24.8	24.1	24.6	24.5	25.4	25.1	24.5	24.8	25.6	24.9	25.7	25.8	26.1	26.3	26.7	26.9	26.6	25.5	25.8	26.1
EFFLUENT-TEMPERATURE	26.6	26.6	26.6	26.6	26.5	26.6	26.6	26.6	26.6	26.6	26.6	27.2	26.6	26.6	27.1	26.9	26.8	27.1	27.4	27.5	27.4	27.2	26.9
INFLUENT-BOD	310	322	342	349	309	290	345	360	302	290	340	310	330	323	360	290	352	338	362	290	298	322	350
EFFLUENT-BOD	114	116	120	124	110	100	112	120	96	96	74	82	88	72	74	58	76	76	72	65	78	82	84
INFLUENT-TSS	550	640	632	640	1120	930	1020	810	440	670	635	911	1170	960	742	706	850	815	730	710	750	1040	950
EFFLUENT-TSS	736	456	476	562	478	406	456	466	554	550	568	741	823	628	782	706	691	633	617	662	655	523	584
INFLUENT-VSS	445	666	498	520	923	756	520	666	366	536	498	756	940	730	466	580	705	665	452	520	595	652	513
EFFLUENT-VSS	396	216	229	285	255	252	246	228	278	274	248	380	430	314	384	352	333	336	306	320	328	261	305

Table E.1 (Con't) Influent vs Effluent – Data Analysis

PARAMETER	6/12	6/16	6/19	6/23	6/26	6/30	6/6	6/9	6/13	6/16	6/20	6/23	6/27	6/30	7/4	7/7	7/11	7/14	7/18	7/21	7/28	7/28	8/1	8/4
INFLUENT-TOTAL PLATE COUNT-AEROBIC	9.50E+07	4.80E+07	6.90E+07	9.50E+07	7.80E+07	1.10E+07	3.20E+07	1.20E+07	1.00E+07	3.20E+07	1.00E+07	4.00E+07	3.20E+07	5.50E+07	7.80E+07	1.10E+07	3.80E+07	3.50E+07	9.50E+07	7.00E+07	5.20E+07	3.20E+07	1.00E+07	1.10E+07
EFFLUENT-TOTAL PLATE COUNT-AEROBIC	1.10E+08	1.00E+08	1.55E+08	7.50E+07	2.00E+08	2.20E+08	1.06E+08	8.60E+07	9.50E+07	1.02E+08	1.06E+08	1.10E+08	1.12E+08	1.12E+08	1.12E+08	8.80E+07	1.06E+08	1.60E+08	1.12E+08	1.14E+08	1.90E+08	7.20E+07	9.20E+07	7.40E+07
INFLUENT-TOTAL PLATE COUNT-ANAEROBIC	6.80E+07	2.20E+07	7.80E+07	6.20E+07	4.80E+07	9.20E+06	2.00E+06	6.40E+06	3.20E+06	6.00E+06	4.50E+06	6.00E+06	5.00E+06	9.00E+06	4.00E+07	8.00E+06	8.00E+06	1.10E+07	4.50E+07	3.80E+07	4.00E+07	1.50E+07	7.80E+06	9.80E+06
EFFLUENT-TOTAL PLATE COUNT-ANAEROBIC	2.20E+05	4.50E+05	3.20E+05	1.34E+06	9.80E+04	6.80E+04	1.02E+05	2.20E+05	2.25E+05	3.80E+05	3.20E+05	7.20E+04	6.80E+04	2.20E+05	2.00E+05	1.00E+05	1.06E+05	6.20E+03	2.20E+05	6.20E+05	6.80E+05	5.60E+05	2.20E+05	2.02E+05
INFLUENT-HYDROGEN SULFIDE PRODUCERS	6.00E+04	5.80E+04	2.10E+04	3.00E+04	5.50E+05	3.00E+06	2.10E+04	6.00E+04	9.00E+04	1.00E+05	6.00E+04	9.20E+05	3.20E+05	4.40E+05	3.00E+05	9.00E+04	7.80E+06	3.00E+05	6.20E+04	9.20E+04	2.10E+04	5.00E+04	3.00E+04	3.50E+04
EFFLUENT-HYDROGEN SULFIDE PRODUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
INFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	1.10E+07	1.10E+07	1.10E+07	4.80E+06	1.10E+07	1.50E+06	2.10E+06	1.50E+06	2.40E+06	2.40E+06	4.80E+06	7.50E+05	1.10E+07	1.10E+07	4.80E+06	1.10E+07	1.10E+07	2.40E+06	1.10E+07	1.10E+07	1.10E+07	4.80E+06	1.10E+07	1.10E+07
EFFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.80E+07	2.30E+07	9.30E+05	2.00E+06	9.30E+06	1.20E+06	2.60E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.50E+05	1.50E+05	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+05	7.50E+05	1.20E+06
INFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	1.10E+07	1.10E+07	1.10E+07	4.80E+06	1.10E+07	1.50E+06	2.10E+06	1.50E+06	2.40E+06	2.40E+06	4.80E+06	7.50E+05	1.10E+07	4.50E+06	4.80E+05	1.10E+07	1.10E+07	2.40E+06	1.10E+07	2.40E+06	1.10E+07	4.80E+06	1.10E+07	1.10E+07
EFFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	2.30E+07	1.50E+07	1.20E+07	2.80E+07	2.30E+07	9.30E+05	2.00E+06	9.30E+06	1.20E+06	2.60E+07	2.30E+06	1.10E+07	4.60E+06	2.30E+06	7.50E+05	1.50E+05	1.10E+07	1.50E+06	7.50E+05	4.30E+06	4.30E+06	2.00E+05	7.50E+05	1.20E+06
INFLUENT-DENITRIFIERS	1.10E+07	2.80E+05	2.80E+05	7.50E+06	9.30E+06	2.10E+06	4.60E+06	1.50E+06	4.30E+05	2.60E+05	2.10E+06	4.60E+06	9.30E+06	1.20E+07	1.50E+07	7.50E+05	9.30E+05	1.50E+07	2.10E+07	4.30E+05	2.60E+05	1.50E+06	9.30E+05	4.30E+05
EFFLUENT-DENITRIFIERS	7.50E+06	2.40E+07	1.20E+07	2.40E+07	1.10E+07	7.50E+06	2.30E+06	2.60E+06	1.10E+06	1.20E+06	9.30E+05	4.60E+06	1.10E+07	2.40E+06	9.30E+04	1.10E+05	2.10E+05	2.40E+06	7.50E+05	2.40E+05	4.60E+06	2.40E+06	1.10E+06	9.30E+05
INFLUENT-SULFATE REDUCERS	1.10E+05	1.50E+04	1.50E+04	1.10E+05	2.40E+04	9.30E+04	1.50E+04	1.50E+04	9.30E+04	4.30E+05	9.30E+05	7.50E+03	1.20E+04	4.60E+05	1.10E+06	2.40E+05	1.50E+05	2.40E+05	1.10E+06	1.10E+06	4.60E+06	2.40E+05	2.40E+05	9.30E+04
EFFLUENT-SULFATE REDUCERS	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01	1.00E+01
INFLUENT-AMMONIA-N									550				430			250				600				
EFFLUENT-AMMONIA-N									389				280			255				330				
INFLUENT-NITRATE									62.0				140.0			34.0				360.0				
EFFLUENT-NITRATE									0.0				0.3			0.0				0.3				
INFLUENT-SULFATE									220.0				122.0			78.4				58.5				
EFFLUENT-SULFATE									0.0				0.0			0.0				0.0				
INFLUENT-SULFIDE																0.8				1.4				
EFFLUENT-SULFIDE									0.0				0.0			0.0				0.0				
INFLUENT-pH	7.45	7.65	7.30	7.21	6.98	6.62	7.11	7.32	7.56	7.84	7.62	7.75	7.21	6.98	6.87	7.14	7.62	7.24	7.81	7.62	7.84	7.75	7.61	7.41
EFFLUENT-pH	8.66	8.78	8.8	8.72	8.8	8.62	8.66	8.62	8.65	8.66	8.75	8.72	8.32	8.56	8.76	9.16	9.13	9.15	9.05	9.1	9.12	9.02	9.05	9.15
INFLUENT-DISSOLVED OXYGEN	0.05	0.05	0.04	0.05	0.78	0.61	0.62	0.88	0.85	0.73	0.81	0.84	0.46	0.04	0.02	0.08	0.05	0.05	0.04	0.04	0.05	0.05	0.04	0.03
EFFLUENT-DISSOLVED OXYGEN	2.2	2.2	2.1	1.98	3	3.35	2.21	2.3	2.2	1.98	2.2	2.5	2.08	3.12	3.19	3.46	3.58	3.2	2.7	2.5	2.28	2.02	2.13	2
INFLUENT-TEMPERATURE	26.4	25.8	25.9	26.5	26.4	26.2	26.5	27.1	27.3	26.2	25.6	24.7	27.3	26.2	27.6	26.0	27.8	26.5	25.6	26.4	26.9	27.5	28.2	27.8
EFFLUENT-TEMPERATURE	27.5	27.9	27.8	27.6	28	27.8	27.5	28.4	29.5	29.4	29.6	30.2	31.2	30	30	30	30	29.5	29.8	29.4	30	30	30.1	29.8
INFLUENT-BOD	330	362	365	344	298	310	360	310	330	280	320	303	300	456	543	460	520	560	430	432	413	362	375	440
EFFLUENT-BOD	78	96	112	74	65	62	68	58	64	50	58	56	45	62	74	68	104	88	62	75	62	60	75	75
INFLUENT-TSS	980	800	560	940	1130	1200	980	802	850	427	526	476	780	919	1162	1210	947	870	960	923	960	1120	780	990
EFFLUENT-TSS	625	654	814	905	969	726	621	657	698	751	960	857	955	926	748	847	847	685	869	723	766	658	675	756
INFLUENT-VSS	784	666	448	525	905	762	720	490	423	345	488	362	625	766	805	1050	770	705	560	410	748	969	642	729
EFFLUENT-VSS	321	338	410	468	434	336	315	328	305	386	422	411	516	289	259	331	219	250	330	336	373	438	473	387

Table E.2 Correlation Coefficient – Influent vs Effluent

PARAMETER	CORRELATION COEFFICIENT
INFLUENT-TOTAL PLATE COUNT-AEROBIC EFFLUENT-TOTAL PLATE COUNT-AEROBIC	0.02
INFLUENT-TOTAL PLATE COUNT-ANAEROBIC EFFLUENT-TOTAL PLATE COUNT-ANAEROBIC	-0.10
INFLUENT-HYDROGEN SULFIDE PRODUCERS EFFLUENT-HYDROGEN SULFIDE PRODUCERS	0.17
INFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS EFFLUENT-CARBOHYDRATE UTILIZERS-ACID PRODUCERS	0.14
INFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS EFFLUENT-CARBOHYDRATE UTILIZERS-GAS PRODUCERS	0.11
INFLUENT-DENITRIFERS EFFLUENT-DENITRIFIERS	-0.10
INFLUENT-SULFATE REDUCERS EFFLUENT-SULFATE REDUCERS	-0.11
INFLUENT-AMMONIA-N EFFLUENT-AMMONIA-N	0.75
INFLUENT-NITRATE EFFLUENT-NITRATE	-0.21
INFLUENT-SULFATE EFFLUENT-SULFATE	0.36
INFLUENT-SULFIDE EFFLUENT-SULFIDE	0.64
INFLUENT-pH EFFLUENT-pH	0.28
INFLUENT-DISSOLVED OXYGEN EFFLUENT-DISSOLVED OXYGEN	0.13
INFLUENT-TEMPERATURE EFFLUENT-TEMPERATURE	0.71
INFLUENT-BOD EFFLUENT-BOD	0.06
INFLUENT-TSS EFFLUENT-TSS	0.02
INFLUENT-VSS EFFLUENT-VSS	-0.07

Table E.3 Influent vs Effluent – Characterization Plots

FIGURES E.1 THROUGH E.16		
E.1	INFLUENT vs EFFLUENT	TOTAL PLATE COUNT - AEROBIC
E.2	INFLUENT vs EFFLUENT	TOTAL PLATE COUNT - ANAEROBIC
E.3	INFLUENT vs EFFLUENT	HYDROGEN-SULFIDE PRODUCERS
E.4	INFLUENT vs EFFLUENT	CARBOHYDRATE-UTILIZERS - ACID PRODUCERS
E.5	INFLUENT vs EFFLUENT	CARBOHYDRATE-UTILIZERS - GAS PRODUCERS
E.6	INFLUENT vs EFFLUENT	DENITRIFIERS
E.7	INFLUENT vs EFFLUENT	SULFATE REDUCERS
E.8	INFLUENT vs EFFLUENT	AMMONIA-NITROGEN
E.9	INFLUENT vs EFFLUENT	NITRATE-NITROGEN
E.10	INFLUENT vs EFFLUENT	SULFATE
E.11	INFLUENT vs EFFLUENT	SULFITE
E.12	INFLUENT vs EFFLUENT	pH
E.13	INFLUENT vs EFFLUENT	DISSOLVED OXYGEN
E.14	INFLUENT vs EFFLUENT	BOD
E.15	INFLUENT vs EFFLUENT	TOTAL SUSPENDED SOLIDS
E.16	INFLUENT vs EFFLUENT	VOLATILE SUSPENDED SOLIDS

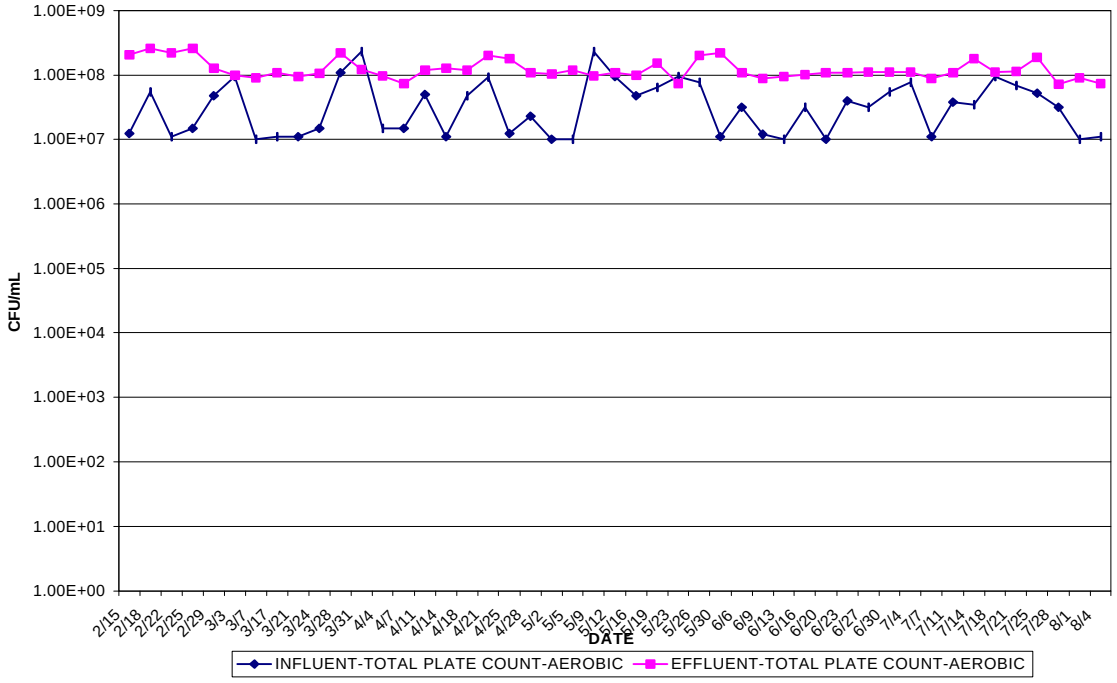

Figure E.1 Influent vs effluent – total plate count - aerobic

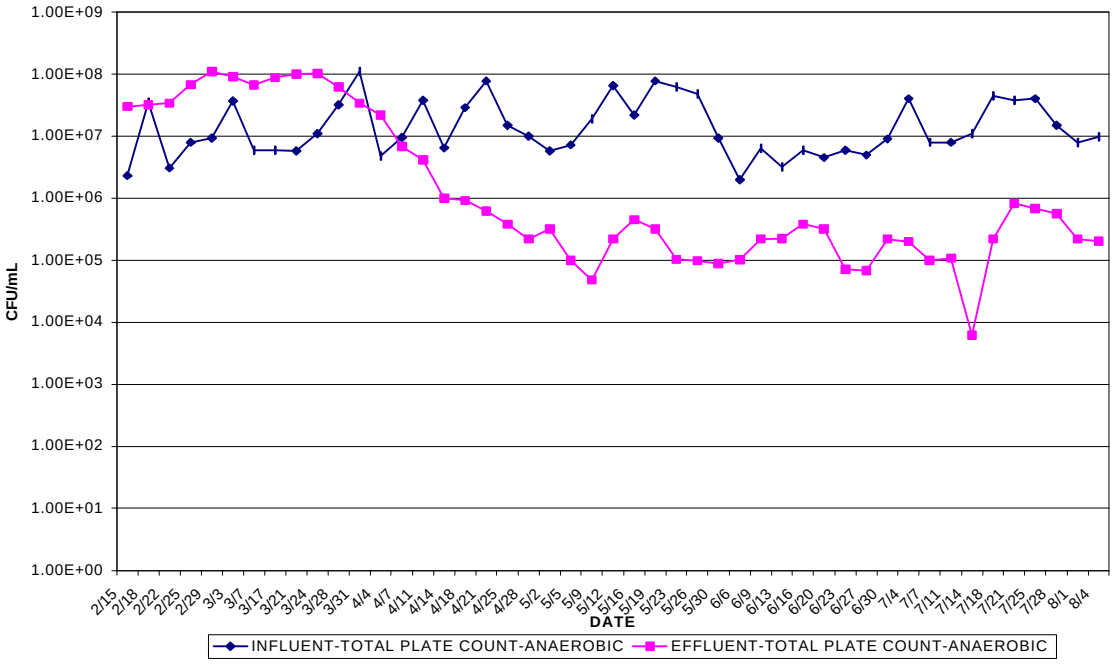

Figure E.2 Influent vs effluent – total plate count - anaerobic

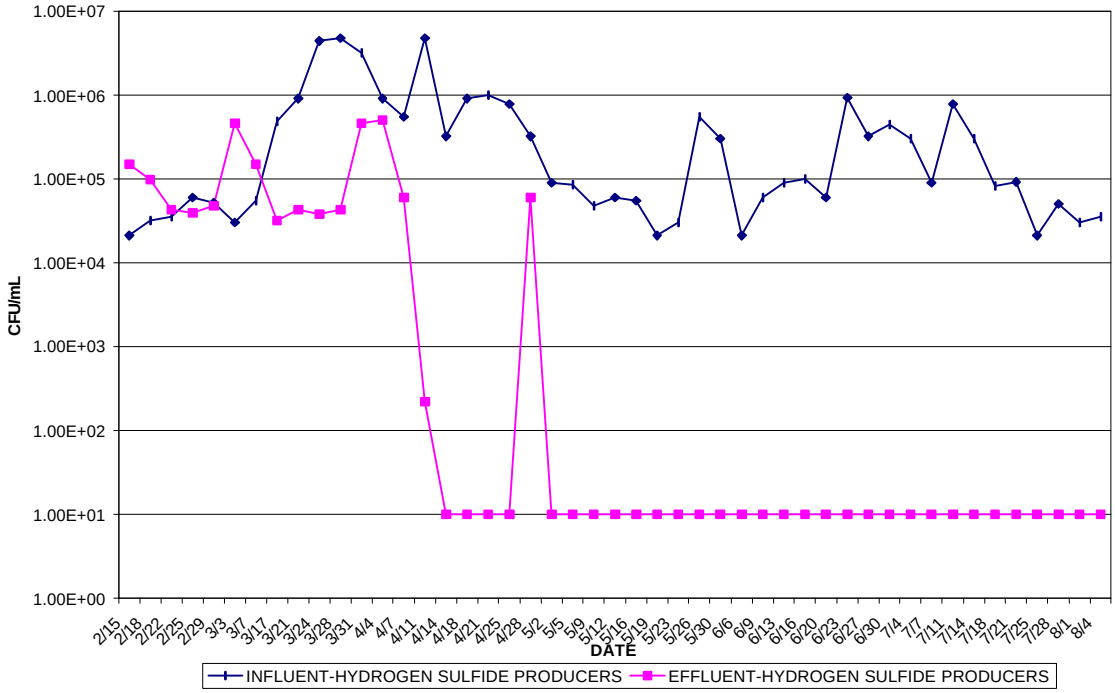

Figure E.3 Influent vs effluent – hydrogen-sulfide producers

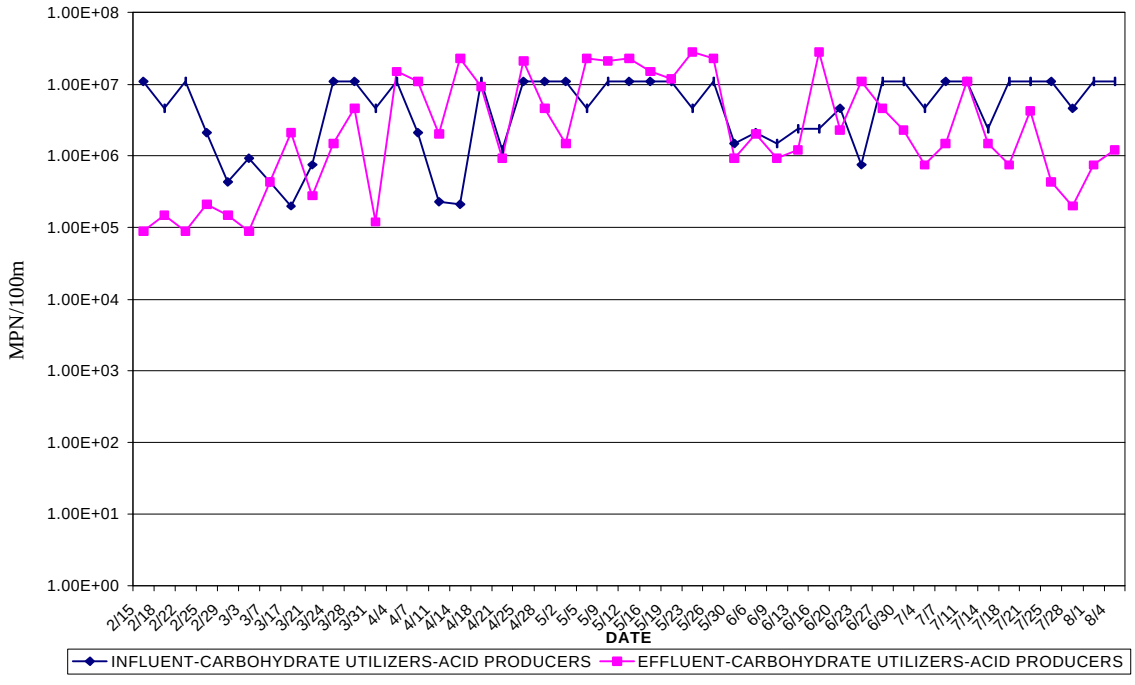

Figure E.4 Influent vs effluent – carbohydrate-utilizers – acid producers

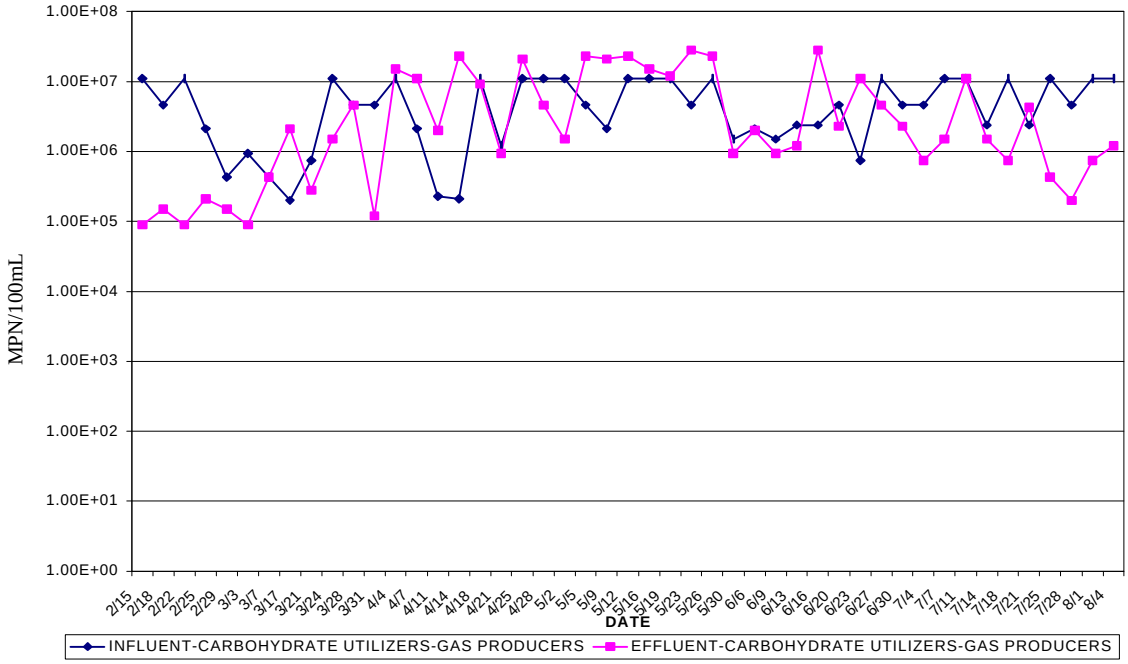

Figure E.5 Influent vs effluent – carbohydrate-utilizers – gas-producers

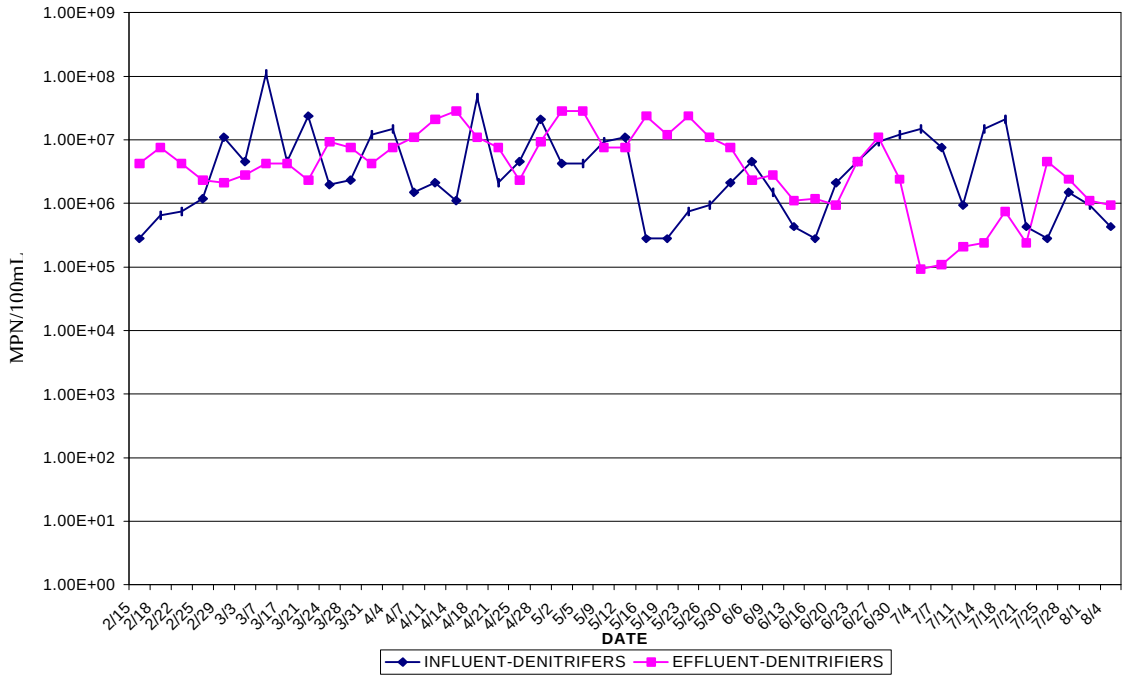

Figure E.6 Influent vs effluent - denitrifiers

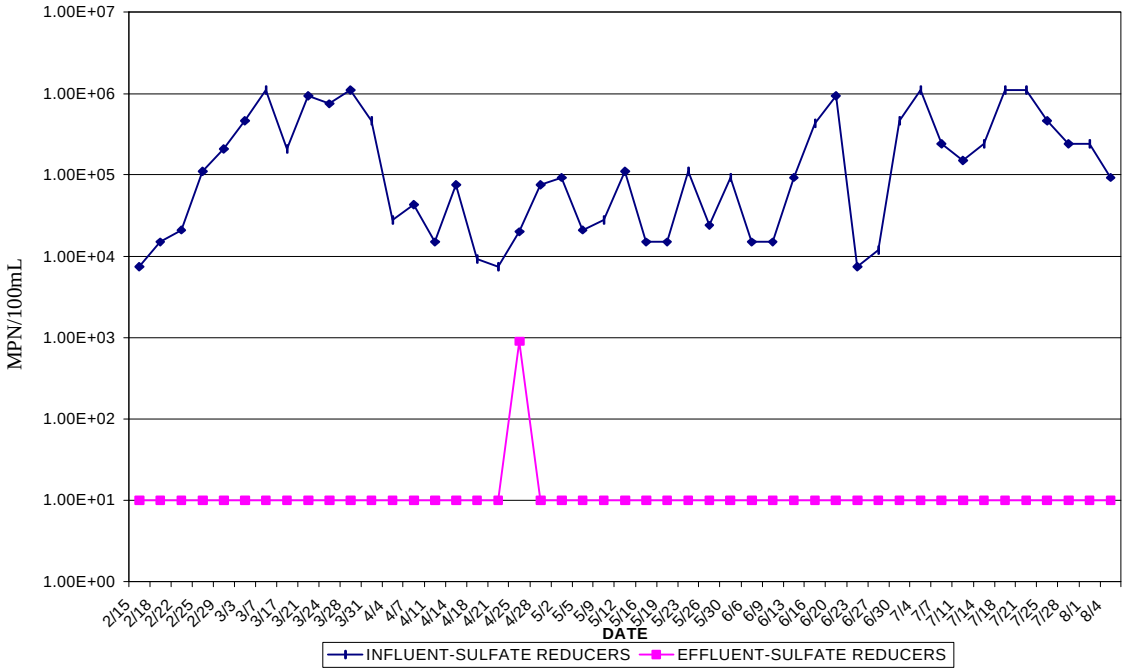

Figure E.7 Influent vs effluent – sulfate reducers

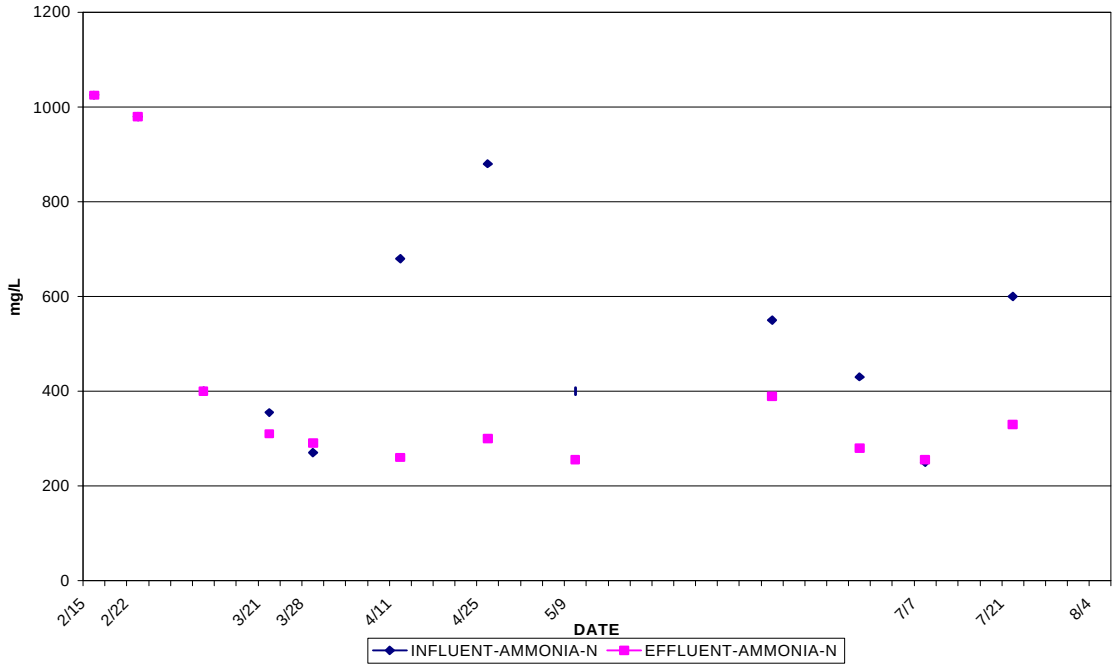

Figure E.8 Influent vs effluent – ammonia-nitrogen

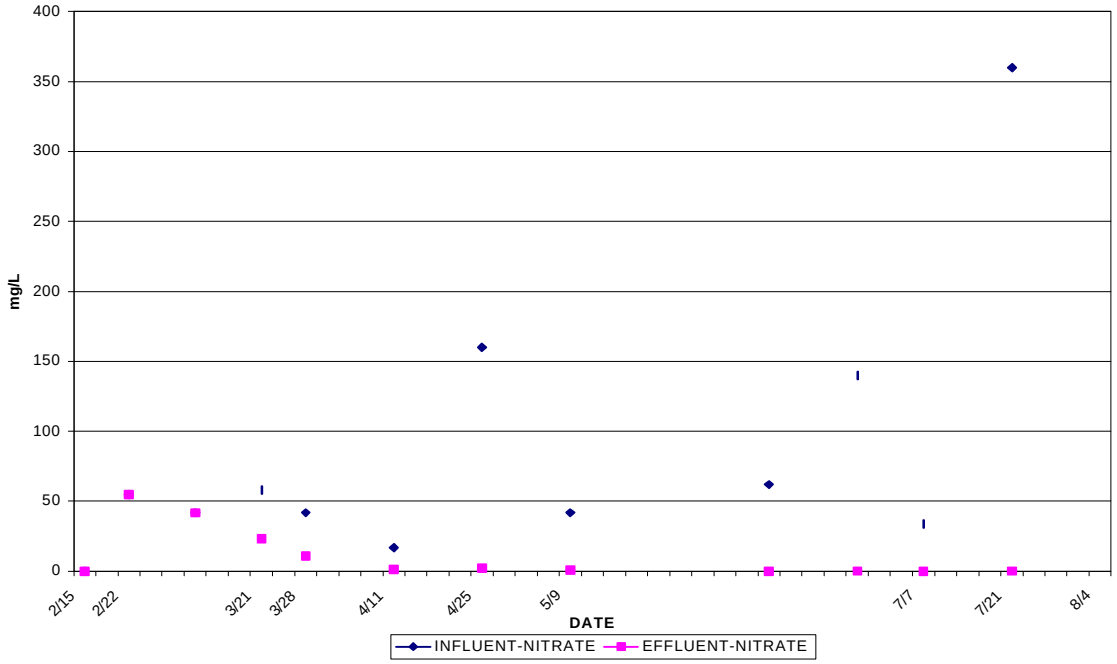

Figure E.9 Influent vs effluent – nitrate-nitrogen

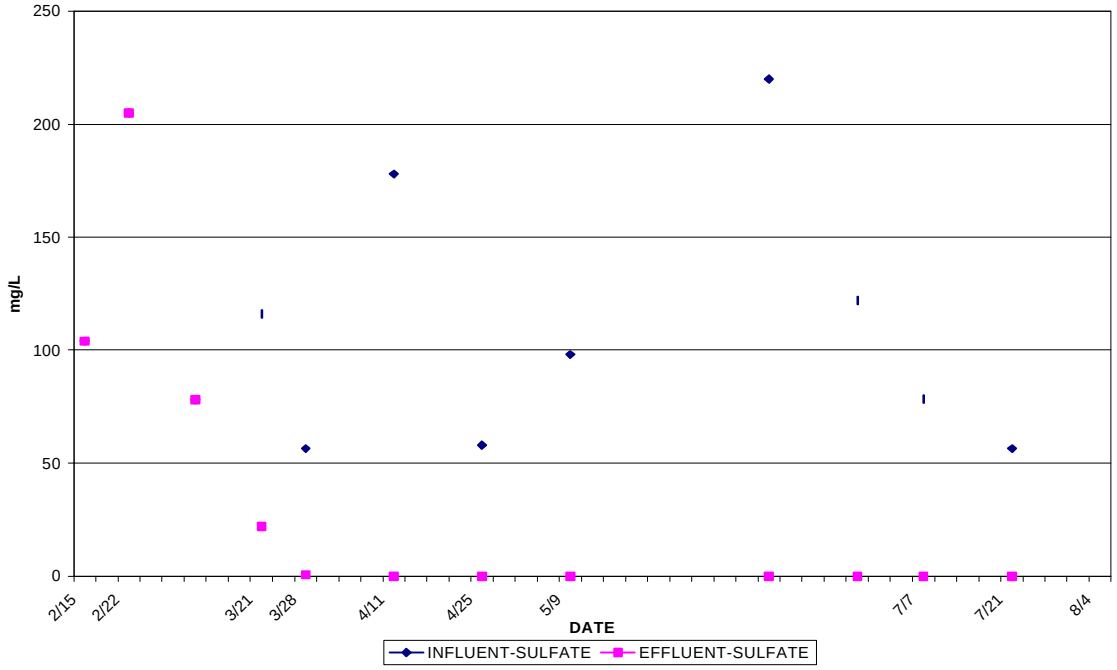

Figure E.10 Influent vs effluent - sulfate

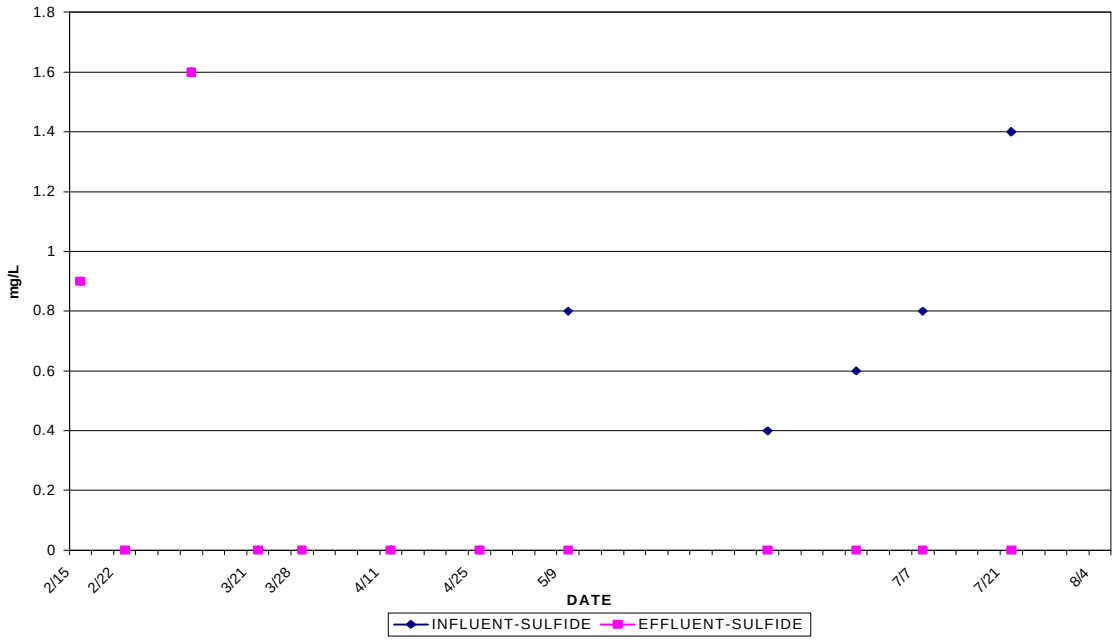

Figure E.11 Influent vs effluent - sulfide

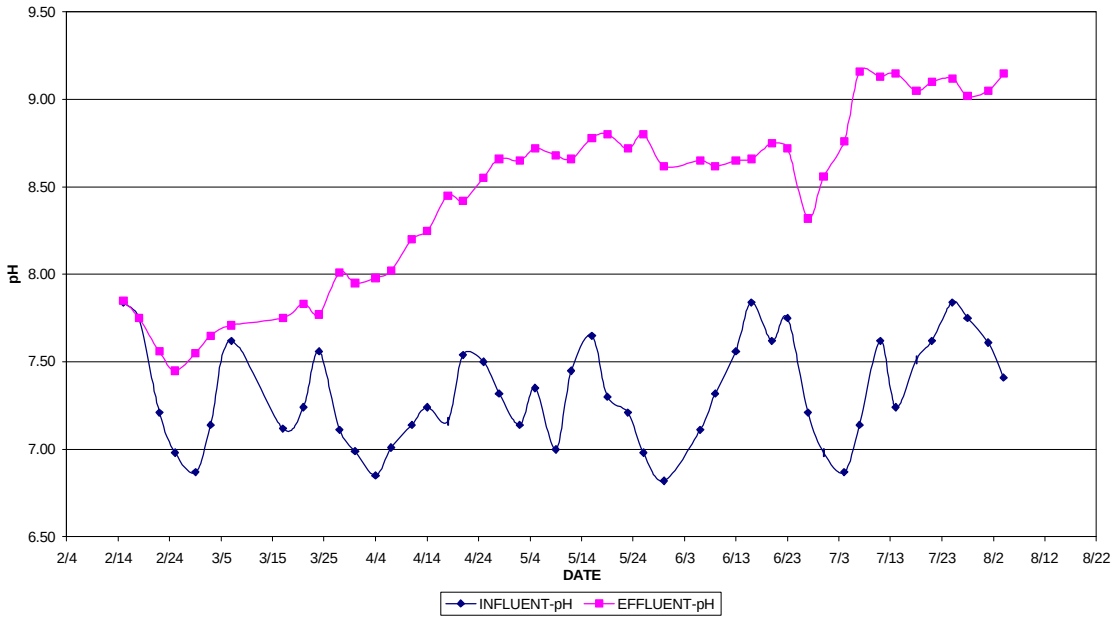

Figure E.12 Influent vs effluent - pH

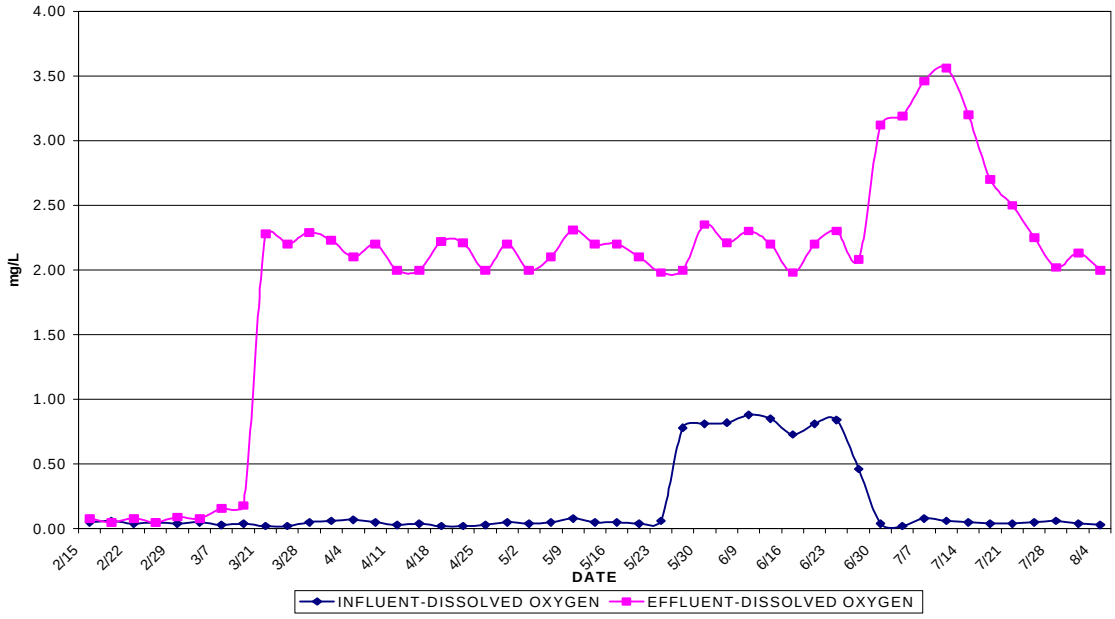

Figure E.13 Influent vs effluent – dissolved oxygen

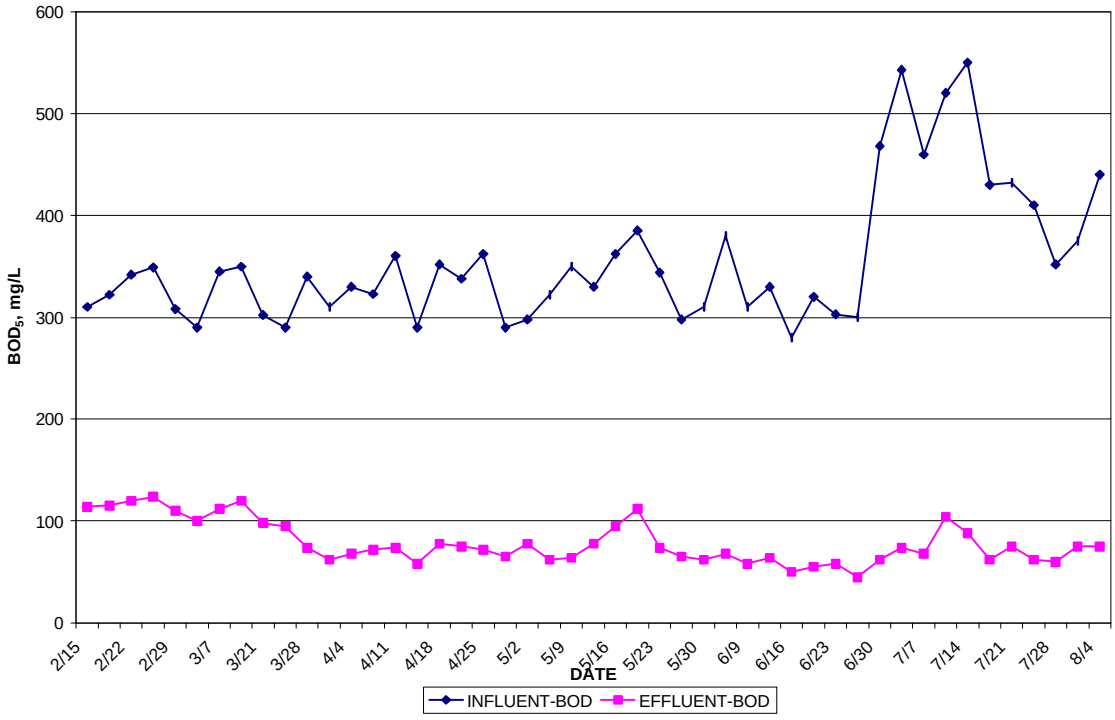

Figure E.14 Influent vs effluent - BOD

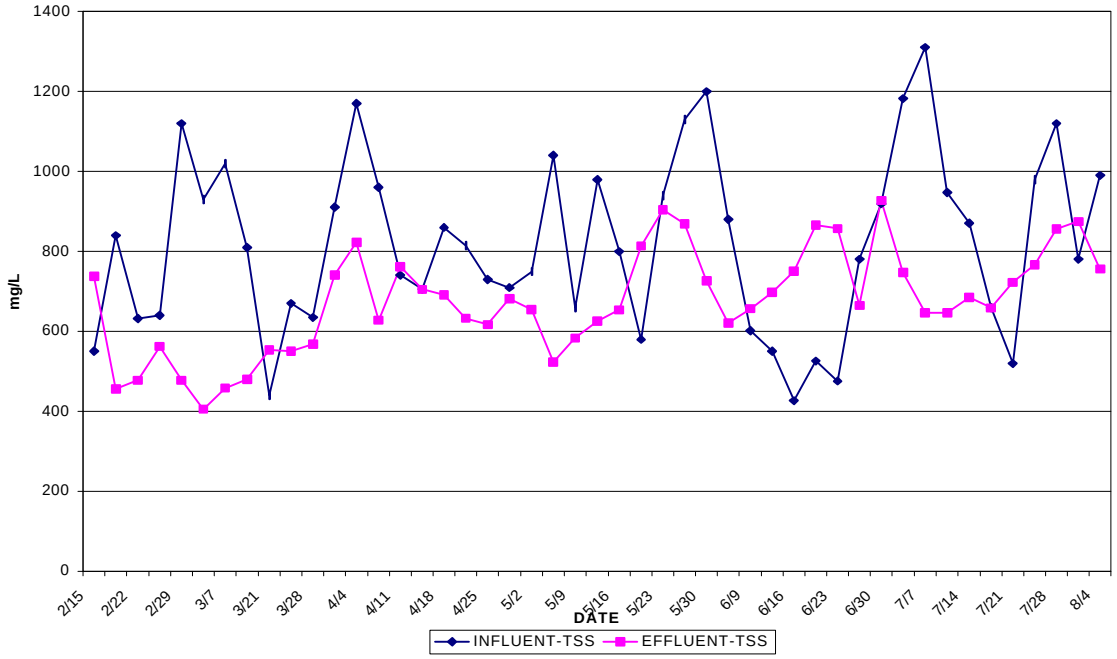

Figure E.15 Influent vs effluent – total suspended solids

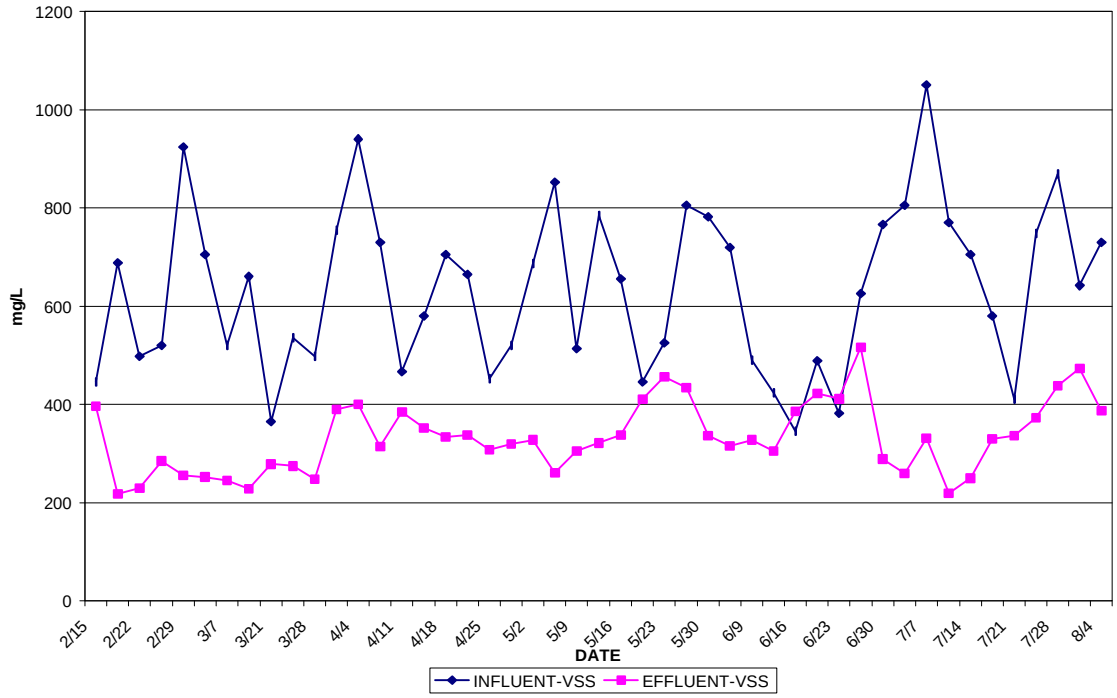

Figure E.16 Influent vs effluent – volatile suspended solids